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Title: GERMINATION, FIELD EMERGENCE AND STORAGE OF NOBLE FIR (ABIES PROCERA REHD.) SEED

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The objectives of this study were to develop a germination test for Abies seeds, which would produce maximum germination in a short testing period; to develop a test to predict noble fir seedling emergence; to determine if certain fungicides were toxic to noble fir seed germination and to determine the optimum temperature and seed moisture content for the storage of noble fir seeds.

The germination responses of 12 seed lots were examined at seven temperatures. Maximum germination occurred at 5° C; however, the germination rate was too slow to be practical for seed testing purposes. Germination rate increased with increasing temperature, but none of the other temperatures consistently produced maximum germination. The increased incidence of molds at the warmer germination temperatures was thought to be related to reduced germination.

Six seed lots were germinated after prechill periods of 0 to 24 weeks. Rate of germination generally increased with increasing lengths of prechill. Lots varied in their prechill requirement and achieved maximum germination following different lengths of prechill. This lot X length of prechill interaction made it impossible to recommend any prechill length for obtaining maximum germination with standard seed laboratory testing procedures.

Soaking seeds in growth regulators for 20 and 40 hours did not result in increased germination. Soaking in water, GA_3, thiourea, and KNO_3 produced higher germination than succinic acid or benzyladenine. Seeds soaked for 20 hours germinated more than seeds soaked for 40 hours. None of the treatments were superior to existing germination methods for germination of noble fir seeds.

The only method which produced maximum germination within a short testing period consisted of chipping the seed coat and germinating at 20-30° C. Removal of 1 to 2 mm from the radicle end of the seed produced maximum germination without prechill in 14 days, thereby shortening the test period by 42 days.

Seedling emergence and survival data for four lots of 1-year-old noble fir seeds were collected by five nurseries. Emergence varied widely among nurseries, demonstrating the impossible task of predicting the percentage seedling emergence for all nurseries. Germination was a more accurate predictor of seedling emergence

than tetrazolium, hydrogen peroxide or excised embryo tests. All laboratory tests, however, successfully ranked the seed lots in order of seedling emergence, but not in terms of absolute percentages.

Seedling losses occurred most rapidly during the first 3 months after sowing, varying from 27 to 71% at different nurseries.

Application of Orthocide 75 to stratified seeds, a common nursery practice, reduced seedling emergence 57%. Arasan 50, Orthocide 75 and Benlate all reduced laboratory germination of noble fir seeds when the fungicides were applied at a maximum adhering rate. Benlate was most toxic followed by Orthocide 75 and Arasan 50. The degree of phytotoxicity was not clearly related to fungicide application before or after stratification. Fungicides were more toxic when applied to non-stratified seeds with a high moisture content (32%) than when applied to the same seeds which were dried to 5% moisture.

Moisture content of two seed lots was adjusted over saturated salt solutions at six levels and the seeds were stored in sealed jars at -18, 5 and 20° C. Germination tests after 0, 6, 12 and 24 months storage indicated that the best overall storage temperature for noble fir was -18° C, but seeds would store equally well at other temperatures if the moisture content was low. Seeds with moisture contents of 12 to 17% retained their viability longest when stored at -18° C;

seeds with moisture contents of 4, 6, 8 and 9% stored equally well at -18° or 5° C; seeds with 4% moisture content stored equally well at all temperatures.

Germination, Field Emergence and Storage of Noble Fir (*Abies procera* Rehd.) Seed

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GERMINATION, FIELD EMERGENCE AND STORAGE OF NOBLE FIR (ABIES PROCERA REHD.) SEED

INTRODUCTION

The true firs belong to the genus Abies, a member of the Pinaceae family. In a recent monograph (Liu, 1971), the firs were described as being widely distributed throughout the Northern Hemisphere. Their utilization includes wood for pulp and timber. Turpentine is also collected from certain species for various commercial uses. In addition, the true firs are very attractive trees and are used as ornamentals in landscapes and as Christmas trees.

In the Pacific Northwest there is an increased demand for the management of Abies. Modern reforestation is becoming oriented more toward transplanting nursery grown seedlings and less toward aerial seeding. Some large scale nursery plantings are being done in containers, rather than in the nursery row. Regardless of planting method, there is an increased emphasis on space utilization which has brought about an increased demand for accurate sowing rates.

Because nursery sowing rates are based on germination percentages, it is important that germination tests be accurate. Concern had been expressed by segments of the tree seed industry regarding the inconsistent germination of Abies seeds. It was not known whether the causes for this inconsistency were due to seed dormancy, sampling variation, germination test conditions, or other reasons.

At the request of the tree seed research committee of the

Western Forest Tree Seed Council, research on the germination of Abies, with emphasis on noble fir, was begun at Oregon State University in 1970.

The original objectives of this study were to (1) develop improved tests for determining viability of Abies seed lots, and (2) develop tests for determining stand establishment potential of Abies seed lots.

As the study progressed, two other areas of work were begun. Reduced seedling emergence related to application of the fungicide Orthocide 75 was observed in the nursery study. Therefore, work was begun to study the adverse effect of Orthocide 75 on the germination of noble fir seeds.

It was learned that the recommended moisture content for the storage of Abies seeds was higher (9-12%) than for other conifer species (6-9%) of the Pacific Northwest. It was felt that there was a need to verify this higher moisture recommendation, which may have been based on research that was no longer valid. This led to a study to determine optimum seed moisture content and temperature for noble fir seed storage.

The four areas of research to be reported on in this thesis, therefore, include studies on (1) viability, (2) seedling emergence, (3) fungicides, and (4) seed storage.

LITERATURE REVIEW

Viability Testing of Tree Seeds

The most common method for determining tree seed viability is the germination test. Other methods are available, however, including the embryo excision test, hydrogen peroxide test, tetrazolium test and x-ray test.

Germination Test

Germination of the true firs is notoriously low. The Woody-Plant Seed Manual (Forest Service, U.S.D.A., 1948) describes the germination of Abies seeds as seldom over 50%. In one study (Rohmeder, 1960) the germination of 71 lots of grand fir varied from 21 to 77%, with a mean of 49%. A review of the germination records of 339 Abies samples tested over a 3-year period at the Oregon State University Seed Laboratory, Corvallis, showed the average germination to be less than 50%.

Many reasons are cited for this low germination. Toumey and Stevens (1928) credited the poor germination performance of A. balsamea to rapid seed deterioration. Empty seeds of grand fir lots were seen to average 32% (Rohmeder, 1960). The Woody-Plant Seed Manual (Forest Service, U.S.D.A., 1948) credits the poor

germination of Abies seeds to embryo dormancy, mechanical injury in seed processing, insect infestations and the perishable nature of the seed. Immaturity is another factor often cited for poor germination (Ching, 1960; Lanquist, 1946; Muller, 1971; Pfister, 1964). Muller recommended that cones of A. nordmanniana be left to disintegrate naturally in store to insure maximum maturity.

Germination inhibitors have also been cited as being responsible for poor germination. Oils present in the seed coat of fir seeds were found to inhibit and delay the germination of pine seeds (Dassler and Zentsch, 1959). In this article, the authors credited Rohmeder with the discovery that the terpine oil present in fir seeds served to prevent premature germination of the seeds which fall to the ground.

Allen and Bientjes (1954) suggested that another reason for low and erratic germination results of conifer seeds might be the lack of reliable testing procedures.

Methods prescribing the procedures for the germination testing of tree seeds were present in the first printing of the International Rules for Seed Testing (International Seed Testing Association, 1953). The number of species was quite limited however. In the United States such official rules were non-existent prior to 1965, at which time the Association of Official Seed Analysts incorporated germination testing methods for tree seeds into their Rules for Testing Seeds (Association of Official Seed Analysts, 1965). Prior to 1965

the Rules for Service Testing Forest Tree Seed of the Pacific Northwest provided guidelines for the germination testing of conifer species (Oregon Agricultural Experiment Station, 1959). A publication entitled Sampling and Service Testing Western Conifer Seeds (Stein, 1966) also provided guidance during this period.

The current methods for testing seeds (Association of Official Seed Analysts, 1970; International Seed Testing Association, 1966) have specific germination recommendations for a large number of conifer species.

The current recommended temperature for germinating Abies seeds is 20-30° C, meaning daily alternations of 16 hours at 20° C plus 8 hours at 30° C. (A constant 25° C is acceptable as an alternate method for some species.) Light is recommended for a minimum of 8 hours, to coincide with the higher temperature of the alternating temperature regime. The intensity of light is to be between 50 and 125 foot-candles and should be of the cool white fluorescent type. Relative humidity levels should be maintained near the 95% level during the germination period. In addition, a prechill period of 14 to 28 days prior to germination is recommended for fresh or dormant seeds. Prechilling conifer seeds consists of exposing imbibed seeds to a temperature of 3 to 5° C for the duration of the prechill period.

Even with the prescribed germination procedures set forth in

these rules, there continue to be problems when testing Abies seeds. Simak (1970a) compared the germination results of silver fir achieved in sand, a Jacobsen apparatus, by the hydrogen peroxide technique, and by the tetrazolium test. He concluded that because of the higher viability indicated by the latter two tests, maximum germination was not being achieved and urged that the causes of the poor germination of Abies seeds be detected and eliminated.

According to Baldwin (1942), Tietz and Kientz were among the first workers to study the effect of temperature on the germination of tree seeds. Later, two American foresters (Siecke and Blumer, 1904) studied the effect of various temperatures on the germination of 24 species of pines. Other quite thorough temperature studies have been conducted on tree seeds (Baldwin, 1934a, 1934b; Ching, 1958; Jensen and Noll, 1959; Kamra and Simak, 1968); however, very few studies have been reported on Abies.

Barton (1930) reported that low temperature stratification improved the germination of A. arizonica seeds. She found that stratification produced more rapid emergence, but the overall production of seedlings was often the same as for unstratified seeds.

Mirov (1936) also reported the beneficial effects of prechilling Abies seeds. He reported a 38% increase in germination when A. concolor seeds were stratified 3 months at 40° F.

Heit and Eliason (1940) found the trend of germination slow and

gradual for laboratory testing of balsam fir, requiring 60 days to complete the test. No germination temperatures were specified. The authors reported that the rate of germination increased considerably, but noted that the germination was only slightly higher, after stratification for 30 days at 2 to 4° C.

MacGillivray (1955) reported that balsam fir seeds could be held in stratification for as long as 14 months without loss of germination, enabling seeds to be held in stratification conditions from one season to another without loss of germination.

Jensen (1964) conducted comparative tests on A. concolor seeds to study the effect of 4 weeks prechilling at 5° C prior to germination. Of 62 lots studied, 34 lots benefited by chilling, averaging 18% higher than the no-chill. Nine lots were injured by chilling, averaging 10% less than the no-chill, and 19 lots were approximately the same under both conditions. The author recommended that both a 4-week chill and a no-chill test be run concurrently when testing A. concolor.

Roe (1941) studied the effect of four temperatures on the germination of balsam fir. He found that alternating 68-86° F was superior to the constant temperatures of 41 to 50° F or the alternating 50-77° F. Later (Roe, 1948) a more extensive study was conducted on the germination of balsam fir. These results again confirmed the optimum germination temperature as being 68-86° F. Both the length and temperature of stratification were studied. He found 41° F

to be superior to a 50° F stratification temperature and noted that a 90-day stratification period was necessary to achieve complete germination within 60 days. All stratification periods between 90 and 240 days gave complete germination in 60 days, but as the stratification period lengthened above 90 days, the rate of germination increased.

Wang (1960) studied three germination temperatures in order to find which was optimum for the germination of grand fir seeds. He found that 25° C produced better germination than 15 to 20° C. His study did not include an alternating temperature.

Low temperature germination of Abies seeds was confirmed when Stein (1951) reported that he had observed noble fir and silver fir seeds germinating on snow. Others have since reported similar incidents (Edwards, 1969; Franklin and Kreuger, 1968).

In a publication reviewing the germination requirements of Abies species (Edwards, 1962) it was recommended that alternating 20-30° C be maintained as the official germination temperature. Further, it was recommended that Abies species should be tested by both chill and no-chill methods, because seed lots will vary in their requirements for chilling.

Embryo Excision Test

As the name implys, embryo excision is a method of evaluating

seed viability on the basis of embryo performance when the embryo is removed from the seed. The purpose for removing the embryo is to overcome one or more factors which may delay seed germination, thereby causing more rapid germination.

Bonnet, who worked with detached bean embryo axes in 1754, has been cited as being the first to employ the technique of embryo excision (Andronescu, 1918).

Harrington and Hite (1923) used the technique to study apple seed germination. They reported that removal of the seed coats caused dormant embryos to exhibit growth.

Crocker and Barton (1931) used the embryo excision technique to study the dormancy of certain rosaceous seeds. They reported that peach embryos germinated well on moist filter paper after being removed from the seed coats.

Flemion (1931, 1933a) worked with excised embryos to study the after-ripening of mountain ash and Rhodotypos kerrioides seeds. She then reported (Flemion, 1933b) that seedlings produced from excised embryos of non-after-ripened Rhodotypos seeds were dwarfed. Dwarf seedlings from non-after-ripened embryos of peach, apple, and hawthorne seeds were also noted by Flemion (1934) although the abnormal growth gradually disappeared.

The recognition of the potential of embryo excision as a rapid test to measure the viability of seeds is generally credited to both

Flemion (1936) and Tukey and Barrett (1936). Tukey and Barrett grew peach embryos on sterilized agar containing nutrients and classified the embryo performance after 7 days into three categories. They found good correlation between viability as indicated by this method and actual field emergence. Flemion felt that her method of incubating peach embryos in peat moss was much simpler in that it did not require the equipment and lengthy preparation time.

Methods for excising embryos of Douglas fir and several pine species were prescribed by Flemion (1938). She reported that seed viability could be determined within 10 days by observing the behavior of excised embryos incubated at 20° C on moist filter paper. Viable embryos were found to exhibit various types of development, while non-viable embryos decayed. Moisture and temperature levels were found to be important. Too much moisture or too high a temperature (above 23° C) were deemed detrimental to the test. In addition, there were good correlations between germination and embryo excision methods for the species studied.

Other more recent papers have broadened the technique to include many species (Flemion, 1948; Heit and Nelson, 1941; Heit, 1943; Heit, 1955). Today the method is recognized by the Association of Official Seed Analysts and several seed laboratories perform this test.

Hydrogen Peroxide Test

The hydrogen peroxide test for seed viability is an accelerated germination test used in testing conifer species. Seeds are first softened by soaking in a 1% solution of hydrogen peroxide for 16 to 24 hours. Then, the radicle end of the seed is cut away exposing the root tip of the embryo. The seeds remain soaking in hydrogen peroxide until the root elongates through the opening at the end of the seed. Seeds with elongated radicles are classed as viable, while those having no visible growth are non-viable.

The discovery that hydrogen peroxide produced a stimulatory effect on seed germination came accidentally from work being conducted on seed disinfectants. Miege (1908) noted that dilute solutions of hydrogen peroxide facilitated the germination of seeds.

An early publication dealing with the role of water in metabolism (Babcock, 1911) also reported a beneficial effect of hydrogen peroxide on the germination of seeds. Babcock reported that corn germinated in hydrogen peroxide was free of mold. He further noted that oxygen required for respiration during germination was provided by the hydrogen peroxide in which the corn seeds were immersed. In addition, Babcock suggested that hydrogen peroxide germination might be used to determine seed viability because he noted that seeds of low germination grew slower in hydrogen peroxide

than seeds of high germination.

According to Massee (1913) two French researchers named Pinoy and Margrou, using hydrogen peroxide as a seed disinfectant, reported a stimulatory effect on the germination of Orobus tuberosus (an old Greek name for members of the Leguminosae family) seeds. They found that seeds immersed in hydrogen peroxide for 5 hours germinated in 8 days, while untreated seeds required a month to germinate.

Massee repeated the work of Pinoy and Margrou on a variety of seeds. He reported that hydrogen peroxide retarded germination. but noted that subsequent seedling growth was rapid.

In a study to evaluate hydrogen peroxide as a legume seed disinfectant, Anderson and Walker (1931) reported that it was not only a good seed sterilant but that it also stimulated seed germination.

Parker and Hill (1955) reported that hydrogen peroxide could be used to determine barley seed viability. They used hydrogen peroxide solutions of 0.03 to 0.06% and were able to evaluate barley viability within 30 hours.

Ching and Parker (1958) developed a laboratory test to measure conifer seed viability using hydrogen peroxide. They found that the viability of several conifer species could be determined in 5 to 9 days after seeds were soaked in 1% solution of hydrogen peroxide following removal of the radicle end of the seed. Comparisons of the

viability percentages obtained by hydrogen peroxide and germination tests showed a high degree of correlation.

Ching (1959) later found that hydrogen peroxide activates the germination of Douglas fir seed by accelerating the early respiration phase of seed germination.

Tetrazolium Test

Two distinct areas of research were involved in the early search for a chemical indicator of seed viability (Anon., 1939). Some researchers worked with organic dyes which stained dead plant tissues without penetrating living tissues. Of these "vital staining" methods, dilute indigo carmine solutions were reported to give the best results. The Russian scientist Neljubow used indigo carmine to stain seeds for observing growth patterns. He is also credited with recognizing that the pattern of stain on the embryo axis was indicative of seed normality and seedling vigor (Moore, 1969). The other area of emphasis centered around "biochemical" methods which depended on the ability of living cells to reduce certain chemicals to colored compounds. The tetrazolium test as it is known today evolved from research in this latter area.

2, 3, 5-triphenyl tetrazolium chloride, often abreviated TTC or T. Z., is a salt which dissolves in water to produce a colorless solution. In the presence of enzymes and live, respiring seeds,

tetrazolium is reduced, forming red triphenyl formazan. It is this pigmented compound which stains the seed tissues and indicates viability.

The history of tetrazolium seed testing is well illustrated in a report by Moore (1969). He describes the search for a chemical stain to detect seed viability as one beginning in 1922 with the discovery, by Turina of Yugoslavia, that living cells in seeds were capable of reducing colorless selenium and tellurium salts to red and black colored compounds. Other persons joined the search including Hasegawa of Japan and Eidmann and Lakon of Germany. It was Lakon, however, who in 1942 began using 2, 3, 5-triphenyl tetrazolium chloride in place of the toxic materials previously used to determine seed viability (Moore, 1969; Porter et al., 1947; Smith, 1951). Since then there have been many papers on the use of tetrazolium in seed testing. An extensive bibliography on the subject appears in the Tetrazolium Testing Handbook (Grabe, 1970).

Lakon's original publication on the use of tetrazolium for seed testing appeared in German as did most of his subsequent papers. For one wishing to review his method, a later paper appeared in English which described his technique for small grains and corn seeds (Lakon, 1949).

In 1950 Lakon published on tetrazolium methods for conifer seeds. Partial reviews of his report (Buszewicz and Holmes, 1957;

Parker, 1953a) indicate a rather long testing procedure (40 hours) and further that Lakon attached great significance to the staining pattern of both the embryo and endosperm. He reasoned that the first symptoms of seed deterioration often occur in the endosperm and then proceded to the embryo.

An extensive program began in 1949 at the Alice Holt Seed Testing Laboratory in Britain to test the validity of tetrazolium testing of 11 conifer species (Buszewicz and Holmes, 1957). The form of tetrazolium used was tetrazolium bromide, refered to under the trade name of "Grodex". The tetrazolium technique employed included: 1. soaking the seeds in water overnight; 2. cutting the seeds longitudinally, slightly off center; 3. soaking the cut seeds in 1% tetrazolium bromide in the dark at 30° C for 3 hours; 4. fixing the stain by soaking the seed in 20% formaldehyde for 15 minutes; 5. extracting and evaluating the embryos. Embryos fully stained and those stained up to 5/6 of the total surface area were counted as viable. There was a good correlation between the tetrazolium tests and germination tests conducted in Copenhagen tanks. Also, contrary to Lakon's view, evaluation of the staining pattern of the embryo alone provided an accurate measure of seed viability.

In 1952 a study was conducted to evaluate tetrazolium chloride as an indicator of the viability of ponderosa pine, western white pine, pinon pine, and Douglas fir (On, 1952). Tetrazolium results were

found to be higher than sand flat germination test results. The difference was attributed to the presence of dead endosperm in seeds with viable embryos, causing the seeds to die in the germination test. After studying the effect of the dead endosperm tissue on seed germination the author established new criteria for evaluating tetrazolium tests enabling a better correlation with germination results.

Parker (1953b) discussed some of the applications and limitations of tetrazolium chloride. He pointed out that color can occur in non-living tissue and is affected by reducing sugars, light and pH.

Grano (1958) found a high correlation between germination and tetrazolium test results for loblolly pine. He used a color index to standardize the interpretation of staining. As suggested by Buszewicz and Holmes (1957), Grano also recommended fixation of the stain to prevent coloration of unstained tissues by photoreduction. He recommended soaking the stained embryos in 15% formalin for 15 minutes. In addition, Grano found that permanent mounts could be made of the stained embryos by spraying them with an acrylic spray.

Simak and Kamra (1963) tested the viability of five samples of Pinus silvestris seeds by x-ray and compared the findings with tetrazolium and germination results. The tetrazolium test was found to be too optimistic when conducted on the embryo only to the exclusion of the endosperm. They felt that in order to properly

evaluate Scotch pine seeds, it was necessary to also look at the endosperm. They found that some seeds with surface necrosis germinated and concluded that the extent of necrosis was important for proper evaluation. Finally, the authors questioned the conclusions of Buszewicz and Holmes when they allowed viable seeds to contain up to 1/6 dead tissue. They felt that the location of the dead tissue would influence the germinability of these seeds.

Peirpoint and Jensen (1964) proposed and evaluated six methods for conducting tetrazolium tests on western white pine seeds. The method they selected was also found adaptable to testing Douglas fir, shore pine and Sitka and Engleman spruces.

Moore (1964) provided a complete list of proven tetrazolium procedures for 39 tree species including guides for evaluation and references for each species.

X-Ray Test

The use of x-rays to measure tree seed quality began in 1903 when a Swedish scientist named Lundstrom used x-rays to study seed setting of forest trees in order to determine if the cones were collectable. He recognized that seeds appearing dark on the x-ray were empty and those which were light were filled (Simak and Gustafsson, 1953). From then until the early 1950's the use of x-rays in seed technology was almost exclusively restricted to the detection of

insects within various types of seeds (Kamra, 1964a).

In a study aimed at exploring the use of x-rays in seed testing, Simak and Gustafsson (1953) developed a method for measuring seed quality using soft x-rays. Seeds were placed on an x-ray film and then irradiated. The resulting x-ray photograph (radiograph) clearly showed the internal seed structure, enabeling various seed quality measurements to be made. For example, the percentage of empty seeds, endosperm development, embryo development, polyembrony, mechanical injury and various other seed abnormalities could be determined. They also studied seed sensitivity to various doses of x-ray irradiation, and concluded that 600 to 1,000 roentgens were lethal to spruce and pine seeds. The dose produced by x-ray photography was not determined, although it was estimated to be 1/10 the lethal dose and was not considered harmful.

The x-ray technique was refined by the development of a system which classified embryo and endosperm tissue (Ehrenberg, Gustafsson, Plym-Forshell, and Simak, 1955; Simak and Gustafsson, 1954). A numerical value was assigned which rated the embryo development from 0, no embryo, to IV, a fully developed embryo. A letter system defined the degree of endosperm development, where A indicated well developed endosperm and B indicated a lack of endosperm development.

Muller-Olsen and Simak (1954) found there existed a high

correlation between embryo and endosperm classes, as determined by x-ray, and the germination of freshly harvested seeds of Scots pine. They said "For practical purposes the germinative capacity of freshly collected seeds of Scots pine may be satisfactorily determined by means of x-ray photography." It was soon learned, however, that such an evaluation was suitable only for freshly collected, physiologically sound seeds. Seeds which had been stored might have a high anatomical potential and germinate very poorly; therefore, some other criterion was needed for an accurate measure of seed viability.

Simak (1957) expanded the usefulness of the x-ray technique when he developed the x-ray contrast method for estimating the germination of Scotch pine seeds. He found that seeds of low viability and seeds with seed coat breaks would absorb barium chloride. Intact, viable seeds would not readily absorb this inorganic compound. Therefore, by soaking seeds in barium chloride before x-raying, Simak was able to produce radiographs which distinguished between viable, injured and non-viable seeds. By combining this contrast technique with the system of embryo and endosperm classification, a more refined technique became available for measuring the germinative capacities of tree seeds.

Two Indian researchers (Swaminathan and Kamra, 1961) then tested the x-ray contrast technique on seeds of 16 non-conifer plant

species and found that they could reliably assess the seed germination potential. The x-ray dose delivered to the seeds was found to be about 15 roentgens and did not adversely affect germination.

Klaehn and Wheeler (1961) used x-rays to study the seed quality of artificial crosses of Norway spruce and white spruce. They found the x-ray technique useful in studying seed quality and reported good correlations between x-ray results and germination.

The importance of selecting the proper contrast agent was demonstrated by Kamra (1963) when he found that organic contrast agents would successfully stain Norway spruce, where previously used inorganic compounds failed.

A new process, described by Brunnekreeft (1963) as contact radiography, consisted of making contact exposures of seeds using soft x-rays. The negative was then enlarged and photographed using an optical microscope.

Simak and Kamra (1963) determined the germinability of Scotch pine seeds by tetrazolium and x-ray contrast, and then compared these results with those obtained by the germination test. They found that the x-ray contrast method was a more reliable indicator of germinability than was tetrazolium. They attributed this to the fact that both endosperm and embryo were considered by the x-ray contrast method, while only the embryo was considered in the tetrazolium method.

Kamra (1964a, 1964b) reported ten separate uses of x-rays in seed testing. Those which related to tree seed testing included the detection of insects, detection of empty and filled seeds, determination of polyembrony, evaluation of embryo and endosperm development, prediction of germination and the detection of mechanical damage.

The viability of Douglas fir seeds was measured by the x-ray contrast method using barium chloride (Sziklai and Hamori-Torok, 1967). These authors found the results to be slightly lower than the actual germination and considered this a positive aspect of the method over other quick tests which were usually higher than germination.

A recent finding showed that weakly developed spruce and pine seeds gave better contrast and differentiation when x-rayed through water (Simak, 1970b).

The current emphasis on container planting for the production of conifer nursery stock has brought greater demands for tree seeds of high viability. It is important that each seed planted produce a seedling. An innovative system developed by Narsted, Nyborg and Sziklai (1972) would select viable conifer seeds on the basis of density using β-radiation in lieu of x-rays. As seed is moved past the sensing unit, viable seeds are allowed to accumulate in one container and non-viable seeds are ejected by an air blast into another container,

thus providing planting stock to meet the increasing demands of the nursery industry.

Nursery Practices

Stratification is a widely used method of overcoming dormancy. It helps insure more rapid germination and uniform emergence of many conifer species (Allen, 1962; Barton, 1930; Edwards, 1962). Originally seeds were mixed with sand and placed in the ground to meet the requirements of stratification (Allen and Bientjes, 1954). This method was practiced by some nurseries as late as 1964 (Issacson, 1972).

Stratification as outlined in the Woody-Plant Seed Manual (Forest Service, U. S. D. A., 1948) consisted of mixing or layering seeds in a container with a wet medium which would retain moisture (i. e., sand or peat moss), then storing the container in a cold room (32 to 41° F) for 30 to 90 days. Following this stratification period the seeds were separated from the stratification medium and planted. Other media, such as vermiculite, began to replace sand and peat moss because it was quite easily separated from the seeds after stratification.

Another method of stratification was devised by Allen and Bientjes (1954) which they called naked stratification. This process consisted of soaking seeds in water for 24 to 30 hours at room

temperature after which the seeds had reached a moisture content of 60 to 70%. Following the soaking period, the seeds were surface dried and then placed in containers with loose fitting lids and stored in refrigerators at 0 to 2° C for several weeks. This practice is now well accepted in forest tree nurseries in the Pacific Northwest.

The Woody-Plant Seed Manual was a widely used reference among forest nursery personnel and served as a guide for much of the currently practiced nursery procedures. It recommended the following nursery practices for Abies: Spring-sown seeds should be stratified 4 to 12 weeks prior to planting, depending on the species. A well-drained sandy loam forms the best seed bed. Seeds should be sown (broadcast or drilled) at a rate to produce 60 to 80 seedlings per square foot. Seeds should be covered with 1/8 to 3/8" of nursery soil and screened from birds and rodents. Abies seeds should be shaded to prevent injury from heat. Today it is common practice to provide heat control by using overhead sprinklers to reduce the surface soil temperatures (Anderson, 1970).

Two disease conditions were recognized as important to Abies seedling production in the Woody-Plant Seed Manual: damping-off and snow mold. The latter disease caused by the fungus Phacidium was reported to occur in Abies balsamea plantings in the Northeast and caused seedling defoliation. Currently, many nurseries commonly treat seeds before sowing to help control a wide variety

of seedling disease causing organisms.

One problem of major concern to many nursery operators has been the lack of correlation between field germination and laboratory germination. Holmsgaard and Kjaer (1951) reported on a study to check the value of germination tests in predicting field emergence of four Abies species. Their results indicated field germination was lower than laboratory germination, but they recognized the value of ranking seed lots by the laboratory germination test. They said "All the species show a clear relationship between germination percent and plant percent, with a lower plant percent for the lower than for the higher germination percent."

Wang (1960) was also concerned with relating laboratory results to field performance. However, he summarized the futility of this when he said "The comparison of nursery germination with laboratory germination is actually not an important one because of the temperature, moisture, and germination medium differences between these two completely distinct environments." He found that one of the two grand fir seed lots studied showed a highly significant correlation coefficient ($r = 0.908$) with a laboratory germination at 20° C, which was a suboptimal germination temperature. No significant correlation was found for the other lot.

Schell (1960) recommended the use of a vigor test to help insure more successful nursery operations. He said that the speed

of germination related to the vigor and size of silver fir and spruce seedlings.

Heit (1969) was also concerned with the poor relationship between laboratory germination and seedling stands in the nursery. He recognized a need for comparative testing to compare laboratory germination results of Abies species with seedling stands in the field. He attributed the lack of desired seedling density in the nursery in part to inaccurate laboratory testing methods but was quick to point out some of the difficulties involved in testing this genus.

Captan and Thiram Toxicity to Conifer Seed Germination

It has been estimated that between 20 and 65% of conifer seedlings die each year after emergence due to pathogenic fungi (Pomerleau and Nadeau, 1959). Therefore, it has become a common practice to treat conifer seeds with various types of chemicals to protect the germinating seedlings from diseases (and other biotic predators) and help insure stands of the desired density. This practice, however, has resulted in reports of phytotoxicity. The literature on the subject is quite extensive and often contradictory.

The early reports involved phytotoxic reactions resulting from the use of acids and salts for controlling damping-off diseases (Hartley, 1915) and for sterilizing tree seeds (Baldwin, 1929; Metcalf, 1925).

As more modern chemicals became available, they were evaluated for their effectiveness in controlling diseases as well as their phytotoxic nature. In such a study, Hamilton and Jackson (1951) reported that fungicide concentrations were important. They found that the germination of shortleaf and loblolly pine was dependent on the concentrations of the various fungicides studied.

Berbee et al. (1953) tested 28 fungicides to determine their effectiveness in preventing damping-off of conifer seedlings caused by Pythium irregulare and Rhizoctonia solani. They reported good control by pelleting seeds with thiram and methyl cellulose as a binder. Concentrations of up to 4 lb. of thiram to 1 lb. of seeds caused no injury in fall sown red pine seedbeds. Vaartaja and Wilner (1956) later attributed this successful control of damping-off to the presence of the additional fungicide bound to the seeds by methyl cellulose, thereby extending its duration of effectiveness.

Vaartaja (1954) reported on a petri dish technique for rapidly testing the effectiveness of chemicals in controlling pathogenic fungi, and their toxicity to germinating seeds. Rhizoctonia and Pythium were used as pathogens, while jack pine (Pinus banksiana) was the host conifer species. Of the 65 chemicals evaluated, most were not effective against the fungi or were toxic to the seeds at effective concentrations. Thiram and captan were reported outstanding, giving good protection at a wide range of concentrations.

An interesting study was conducted on spruce and caragana seeds (Cram and Vaartaja, 1955) which showed that phytotoxicity occurred most often when the fungicide was applied after the seeds had been stratified. Eight different pesticides were applied to the seeds by agitation, and the excess screened off, resulting in a maximum adhering dosage. One-half of the seeds were treated before stratification, the remaining half were treated after stratification. The phytotoxicity of the fungicides was measured by speed of germination and total germination. Fungicides applied before stratification had the least harmful effect on germination, while those applied after stratification often caused severe reductions in germination. Orthocide 75 (75% captan) was listed as mildly toxic in this study, yet the germination of Colorado spruce (Picea pungens) treated after stratification was 63% compared to 98% when treated before stratification.

Another study (Jacks, 1956) which evaluated the effectiveness of several seed dressings in controlling damping-off of Monterey pine seeds, reported that Orthocide 75, at a concentration of 17%, effectively controlled damping-off caused by Pythium and Pellicularia, and was not phytotoxic. (It should be noted that no stratification treatments were included in this experiment.)

By the middle 1950's the practice of pelleting conifer seeds with methyl cellulose, fungicides, and various other materials was

being explored. Reports varied regarding the effectiveness of disease control and phytotoxicity (Kahler, 1955; Shea, 1959; Shea, 1961; Vaartaja and Wilner, 1956; Weihing et al., 1961). One author Vaartaja, 1955) reported that heavy cellulose coatings may reduce germination, but the addition of a fungicide would lessen this effect.

A thorough review of the entire problem of controlling diseases in conifer nurseries was presented by Vaartaja (1964). This work includes 662 references and a section devoted entirely to seed treatments. In summarizing the current research concerning the use of seed treatments, Vaartaja said that even the least phytotoxic materials, such as captan and thiram, would decrease germination if used in high concentrations. He said that conifers required the sustained presence of fungicides for effective control of damping-off, and felt that seed pelleting helped retain the fungicide longer by providing a reservoir. In addition, he speculated that methyl cellulose might also protect the seeds from fungicide toxicity.

Studies by Shea (1959, 1961) suggested that phytotoxicity might not be entirely the result of the fungicide material, but rather, may vary depending on the formulation of the fungicide. Another paper (Carlson, 1970) supporting this view cited an example where a 90% active ingredient formulation of captan was far less phytotoxic than a 50% formulation. Carlson said that additional research on the phytotoxic effects of inert ingredients was needed before continuing

a program of screening seed-treatment chemicals.

Several authors reported that individual conifer species react differently to fungicides (Carlson, 1970; Dobbs, 1971; Weihing et al., 1961). Dobbs, for instance, found thiram to be phytotoxic to white spruce seeds, but not to jack pine.

Lungescu and Manu (1963) reported that several fungicides, including captan and thiram, reduced the respiration and germinative capacity of spruce seeds at concentrations above 1%.

An interesting hypothesis was proposed (Vaartaja et al., 1964) for the inconsistent control of conifer seedling damping-off. In their hypothesis of biological control, they suggested that fungicides may exert a double action: one directly against the pathogen; the other, biologically, by controlling or modifying antagonistic soil flora. It is the dynamic character of soil flora which they said was responsible for inconsistency. They further stated that until soil microbiology was further advanced one must be ready to accept a certain amount of unpredictability from even the best fungicides.

Cayford and Waldron (1967) studied the effects of Captan 50W on the germination of white spruce, jack and red pine seeds. They found that this material, when applied at a rate of 12%, was phytotoxic when the pelleted seeds were surface-sown. When sown beneath the soil, however, the number of abnormal seedlings decreased. Phytotoxic effects were observed as injury to the seedling

roots. They concluded that satisfactory germination could be achieved using captan-treated seeds if the seeds were sown beneath the soil.

Jorgensen (1968) compared the laboratory germination with field germination of Arasan 42-S (thiram) treated longleaf pine seeds. He found that the laboratory germination decreased with increasing concentrations of this fungicide, while germination in the field was not dependent upon the fungicide concentration. He concluded that under field conditions the phytotoxicity of Arasan 42-S was reduced to an insignificant level. A similar conclusion was reached by Demeritt and Hocker (1970).

Carlson and Belcher (1969) tested 61 seed treatment chemicals to determine their effect on the germination of jack pine, lodgepole pine and white spruce. Both captan and arasan (thiram) were found to inhibit the germination of all three species. These authors questioned the continued use of these two materials for control of conifer seedling damping-off.

Peterson (1970) found that captan, and most of the fungicides commonly used as seed protectants, adversely influenced both the amount and speed of germination of ponderosa pine seeds. In addition, he reported that high temperatures would compound this adverse effect.

Bloomberg and Trelawny (1970) further studied the effect of

thiram on the germination of Douglas fir seeds. They reported that germination was delayed if seeds were treated before stratification rather than after. This is in direct contrast to Cram and Vaartaja's 1955 study on spruce and caragana seeds. Another observation was that germination reduction did not occur in seeds with high viability. This may account for some of the seemingly conflicting data in the literature.

Storage of Conifer Seeds

Cone production of noble fir, like that of most conifers, is cyclical. According to the Woody-Plant Seed Manual (Forest Service, U.S.D.A., 1948), most species of Abies produce good seed crops every 2 to 4 years, while the production of A. procera is described as "infrequent". The sporadic nature of cone production, coupled with the yearly demands for quality seed stock, has placed much importance upon the storage of conifer seeds.

Much of the early work regarding the optimum storage conditions for conifer seeds was conducted by European workers. Cieslar, whose work on Norway spruce, black (Austrian), and white pine was published in 1897, is often cited as a pioneer in conifer seed storage research (Barton, 1951; Tillotson, 1921). He concluded that air-tight storage lengthened the life of these species and preserved their germinating power. Haack is another frequently cited early

worker in this area of research (Anon., 1928; Barton, 1961; Tillotson, 1921). In 1909, Haack, reporting on his work with Scotch pine seeds, also found sealed storage better than unsealed storage for maintaining seed viability. Haack also noted the importance of drying seeds prior to storage.

Tillotson (1921) reported on the storage of ponderosa pine, western white pine, eastern white pine, Engelman spruce, Douglas fir and lodgepole pine. Seeds of these six species were dried for 2 days by means of a fan. The seeds were then divided among five kinds of storage containers and shipped to 13 geographical locations throughout the United States to be stored for 5 years under three temperature regimes. Germination tests conducted yearly indicated that air-tight containers were far superior to any other container type tested for maintaining viability. Tillotson also noted that seeds stored in air-tight containers were not affected by temperature differences. Unlike other researchers, Tillotson reported that indoor temperatures were superior to either the fluctuating temperature of an outbuilding or the low temperature conditions of a cellar. The low temperature of his experiment, however, was above the range of 0 to 32° F that was shown to be superior by other workers.

An anonymous article (Anon., 1928) concluded that cold storage temperatures of 9 to 14° F were beneficial for the storage of noble fir seeds over a 5 year period. Isaac (1934) reported further on the

study appearing in the anonymous article. In two separate tests, seeds of noble fir were stored for 5 years at room temperature and 15° F. Each of these two tests resulted in similar results. After 1 year at room temperature the germination had reached zero, but after 5 years at 15° F, the germination had only slightly decreased. Isaac reported that storage at 15° F not only maintained viability, but also improved germination. In both tests germination increased to its highest point after 4 years' storage.

By the 1930's, then, the available information suggested that low temperature storage, as low as 9° F, was beneficial for maintaining conifer seed viability.

Barton (1935) further quantified temperature, as well as studied the effect of sealing, vacuum and desiccation on conifer seed storage. She found that sealed storage and temperatures as low as -15° C were beneficial for maintaining viability of conifer seeds in storage. A vacuum condition, in which the air was exhausted from the storage flasks, helped preserve viability of seeds stored at room temperature. The use of a vacuum, however, for maintaining the viability of seeds stored below freezing temperature was thought unnecessary. Desiccation was achieved either by placing the seeds over calcium oxide or by mixing varying amounts of quicklime with the seeds before sealing. Some harmful effects from extreme drying were reported.

A study (Barton, 1941), consisting mostly of non-conifer species,

was one of the first to examine the effects of both seed moisture and temperature on seed viability. Several aspects of seed storage were considered. Longleaf pine (Pinus palustris) seeds were placed in humidity chambers at 35, 55 and 76% relative humidity and stored at 5, 10, 20 and 30° C for up to 372 days. It was found that viability was maintained better at 5° C than at any of the other temperatures studied. Seeds stored under the high relative humidity of 76% even stored quite well at 5° C for up to 232 days. At temperatures above 5° C, the seeds stored at the higher humidities did not maintain their viability as well as those stored at the lower humidity condition. In addition to the viability aspect of storage, this paper also examined the water absorbtion pattern of various kinds of seeds under differing humidity and temperature conditions.

Barton (1953) included Abies seeds for the first time in one of her storage studies. Seeds of grand fir (A. grandis), noble fir (A. procera), and Shasta red fir (A. magnifica var shastensis) were adjusted to moisture contents ranging from 7 to 18% by placing them over sulfuric acid solutions for 6 weeks. They were then stored in sealed containers at two temperatures, room and a low temperature which was described as 8° C for the first 9 years followed by -4° C for the remaining 7 years. Most of the seeds stored at room temperature died within the first year of storage, while cold temperature storage was more effective at maintaining viability. All species

showed significant germination after 16 years or cold temperature storage. In this same paper, Barton reported that low and high seed moisture content were both harmful to seed germination. In her words, "This was especially noticeable for A. grandis where lowering the moisture to 9% or raising it to 12 or 15% decreased germination from the 98% exhibited by seeds with 11% moisture to 54 and 64%." (Refer to the data below which represent part of a table appearing in Barton's 1953 paper.)

Species	Moisture Content	Percent seedling production after storage for years								
	%	0	1	2	9	10	11	14	15	16
A. grandis	9	58	44	27	27	34	24	19	2	17
	11	98	24	15	57	45	46	36	3	26
	12	54	38	2	9	29	22	4	0	6
	15	64	36	13	19	14	15	9	5	8

Schubert (1954) reported on the viability of various conifers after storage at 5° C for 2 to 24 years. The viability of five species of Abies was characterized as declining quite rapidly (within 3 to 6 years) when compared with other species.

Now that subfreezing temperatures were becoming more common for the storage of conifer seeds, Barton (1954a) further established their superiority and defined more clearly the optimum

storage temperature. She studied the effect of -4, -11 and -18° C on the viability of ponderosa pine, Douglas fir, Sitka spruce, western red cedar and western hemlock. However, her study lacked controlled moisture conditions since the seeds were stored in canvas bags. Of the three subfreezing temperatures studied, it was found that all seeds except ponderosa pine kept better at -18° C than at -11 or -4° C. Deterioration was most rapid at -4° C for all species except for ponderosa pine which stored better at -4° C than either of the other two temperatures.

Barton (1954b) conducted a second study in which she examined the effects of both seed moisture content and storage temperature on seed viability. And, although limited to two levels of seed moisture and two levels of storage temperature, it provided information which should have aleviated some of the skepticism which then existed regarding the use of so-called "frozen" seeds. In the study, Douglas fir seeds in sealed storage with moistures of 5.8 and 13.6% stored equally well at -18° C for 2 years. When stored under the same conditions at 5° C, seeds with 13.6% moisture deteriorated more rapidly than the drier seeds. Seeds were then stored in sealed tin cans, foil envelopes and manila envelopes at 5° C and 30° C. The effectiveness of the container type varied depending on seed moisture content. It was also found that 24 months of storage at -18° C had no harmful effects on seeds when packaged and stored for an

additional 6 months. In fact, the seeds stored at -18° C were found to be more resistant to subsequent adverse storage conditions than seeds stored at 5° C.

Allen (1957) studied 11 species of conifer seeds and found that, in most cases, storage temperatures of 0° F and 32° F maintained seed viability equally well over a period of 5 to 7 years. He also concluded that the seed moisture content did not reduce viability in storage. In examining his data, however, seed moisture was seen to be confined to normal limits with the low moisture 6.3 and the high moisture 9.7%.

Jones (1962) recommended that noble fir be stored at seed moisture contents between 5 and 8% at 20° F. In the same year, Holmes and Buszewicz (1962) reported that noble fir stored well at moisture contents of 6 to 8% in sealed containers, regardless of the storage temperature. However, they noted deterioration when seeds with 16% moisture content were stored at the temperature extremes of 20 and -20° C.

MATERIALS AND METHODS

Seed Lots

When this study began in 1970, the only seed lots available for study were lots harvested in 1968 and stored at -18°C for approximately 1 year. Initial work was conducted on six of these lots. In 1971, six freshly harvested lots were secured for duplicate study. As the seed lots were obtained, viability data were collected. This information is seen in Table 1.

All 1968 seeds were commercially collected and processed; nothing was known of their harvesting or processing history. The remaining six lots, however, were obtained as cones and the care they received in harvesting and processing is known. The cone collection dates are shown in Table 1. Lots 7 and 8 were stored in 2-bushel, open weave grain sacks on covered, open-air drying racks. These cones opened in the bag and did not require forced air drying. The cones of lots 9, 11 and 12 were hand harvested from standing trees in early September and were transported to the laboratory within 1 week. They opened when dried with ambient forced air. The cones of lot 10 were collected from a squirrel sampling and also opened when dried with unheated forced air.

The seeds were separated from the open cones by tumbling

Table 1. Description of seed lots.

Lot	Kind of seed	% Germination		% Viability			% Filled[c] seeds	Collection date	Additional comments
		Chill	No chill	T. Z.[a]	H_2O_2[b]	Excised embryo			
1	Noble fir	87	80	85	93	91	98	1968	Commercially collected & processed
2	Noble fir	7	0	6	21	28	76	1968	Commercially collected & processed
3	Noble fir	15	1	12	21	23	54	1968	USFS collected & processed
4	Noble fir	0	1	5	26	2	58	1968	USFS collected & processed
5	Noble fir	30	45	42	57	33	71	1968	Commercially collected & processed
6	Noble fir	21	13	33	57	--	70	1968	Commercially collected & processed
7	Noble fir	54	52	56	51	50	70	9/29/70	Commercial collection-OSU processed
8	Noble fir	47	53	48	52	46	53	10/13/70	Commercial collection-OSU processed
9	Grand fir	71	64	48	--	--	--	9/9/70	Single tree-OSU processed
10	Noble fir	29	14	32	8	--	38	9/16/70	Squirrel cut
11	Noble fir	12	18	22	14	14	15	9/9/70	Single tree--low elevation
12	Silver fir	20	9	32	2	--	31	9/10/70	Composite of 2 trees-OSU processed

[a] Tetrazolium [b] Hydrogen peroxide [c] Determined by x-ray

the cones in a small wire mesh cage which allowed the seeds to pass through while the cone parts were retained. The seeds were then dewinged by hand rubbing and cleaned on a table model two-screen motor-driven Clipper seed cleaner. The top screen was a 10/64 x 3/4 slot, the bottom screen was a #8 round. Air was maximum for the machine.

All seeds were stored in sealed gallon containers at 5° C until ready for use.

All seed lots received a final cleaning prior to germination by blowing for 1 minute in a South Dakota Seed Blower at a gate opening of 32. Additional heavy inert matter such as pitch and cone parts was also removed by hand.

Germination Procedures

Germination tests were conducted in covered clear plastic dishes measuring 4-5/8 x 4-5/8 x 1-1/8 inches. The dishes were partially filled with 200 ml of sponge-rok[1/] which was moistened by evenly distributing 40 ml of tap water over the medium. Fifty seeds were then placed on top of the medium in each dish with the aid of a vacuum planter. The number of 50-seed replicates comprising a

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[1/] Sponge-rok is the trade name of an exploded rock product manufactured by Paramount Perlite Co., 16236 South Illinois St., Paramount, California 90723.

germination test varied from three to four, depending on the experiment.

After the seeds were planted the dishes were placed in rows of three or four on trays and allowed to germinate at the desired temperature. Alternating temperatures of 20-30 and 5-30° C consisted of 16 hours at the lower temperature, followed by 8 hours at the warmer temperature on a daily alternating cycle. Temperature was controlled to within ± 1° C.

When seeds were germinated in light, it was provided for 8 hours during the period of high temperature. When seeds were germinated in the dark to exclude any effect of light on germination, each germination dish was wrapped in aluminum foil.

Germination counts were made at 7-day intervals. A seed was counted as germinated when the radicle length was equal to the length of the seed. The tests were terminated when maximum germination had occurred.

Methods for Improving the Laboratory Germination of Noble Fir Seeds

Germination Temperature

Effect of Temperature on Total Germination. This study was conducted to determine the optimum temperature(s) for the laboratory germination of *Abies* seeds.

Each of the 12 lots was germinated with four replicates of 50-seeds in the dark at temperatures of 5, 15, 20, 25, 30, 5-30 and 20-30° C.

The duration of the germination tests varied considerably depending on the temperature. Excessive mold and decay often became the criteria for terminating a test.

Effect of Temperature on Rate of Germination. The germination rate (GR) as used by Maguire (1962) and others was calculated for each lot at each of the seven germination temperatures described in the preceeding study.

Germination rate as used here is defined as the summation of the quotients produced by dividing the number of new germinants each week by the number of weeks at which the count was made. Arithmetically, it can be expressed:

$$GR = \frac{\text{number of germinants}}{\text{week 1}} + \ldots + \frac{\text{number of germinants}}{\text{final week}}$$

Both the GR values and the germination percentages were analyzed by a two-factor analysis of variance. Factor 1 was lots, with 12 levels. Factor 2 was germination temperature, with 7 levels.

Length of Prechill

Seeds from noble fir lots 1 through 6 were subjected to prechill

treatments to determine the optimum length of prechill for maximum laboratory germination.

Prechill treatments consisted of placing 50 seeds, in replicates of four, on top of moistened sponge-rok within covered plastic germination dishes and storing at 5° C. Every 2 weeks for 24 weeks a set of four dishes was removed and placed in a 20-30° C germinator with light. Germination counts were conducted every 7 days. After 42 days the germination tests were concluded and all remaining seeds were cut with a razor blade to determine the number of firm ungerminated seeds.

The germination results were analyzed using a two-factor analysis of variance. Factor 1 was lots, with 6 levels. Factor 2 was length of prechill, with 13 levels.

Growth Regulators

Sixteen groups of 50 seeds of lot 1 (Table 1) were soaked in the following solutions: 200 ppm GA_3, 200 ppm thiourea, 5.6 ppm benzyladenine, 16.8 ppm succinic acid, 200 ppm KNO_3 and tap water.

The seeds were soaked in a dark germinator at 25° C for 20 and 40 hours, after which they were removed from the solutions and planted on sponge-rok. Four replicates of 50 seeds from each treatment were germinated in the dark at 15 and 5-30° C without prechilling.

Germination counts were made every 7 days and the tests were concluded after 49 days.

The germination results were analyzed using a three-factor analysis of variance. Factor 1 was the growth regulator soak solution, with 6 levels. Factor 2 was length of soak, with 2 levels. Factor 3 was germination temperature, with 2 levels.

Seed Coat Chipping

Single edge razor blades were used to remove 1 to 2 mm from the radicle end of dry seeds of lot 1 (Table 1). Four replicates of 50 seeds were then germinated without prechilling at 20-30° C with light. Weekly germination counts were made and the tests were concluded after 35 days. A check test was conducted without removing the radicle end of the seed.

Noble Fir Seedling Emergence and Survival

Plantings were made at each of the following five nurseries: Forest Research Laboratory, Corvallis, Oregon; Vancouver Island Nurseries, Duncan, B. C., Canada; Webster Nursery, Olympia Washington; Ben Lomond State Forest Nursery, Santa Cruz, California; and the Dwight L. Phipps Nursery at Elkton, Oregon.

Each nursery received samples from four lots of seed (lots 1, 2, 3 and 5 described in Table 1). Each sample consisted of eight

packets containing 100 seeds each. Seeds in four of the packets were treated with Orthocide 75,[2/] while seeds in four packets remained untreated. A planting plan was provided and each nursery was asked to plant according to it. A randomized block design, consisting of four lots, two treatments, and four replicates was arranged within a plot measuring 6 x 30 feet. Seedling emergence data collected from all nurseries were analyzed as a completely randomized design, factorial arrangement of treatments. Factor 1 was fungicides, consisting of 2 levels. Factor 2 was lots, consisting of 4 levels. Factor 3 was nurseries, consisting of 5 levels. No statistical treatment was attempted for seedling survival data.

Before sending to the nurseries, each 100-seed packet was x-rayed and the number of filled seeds was determined. A Faxitron 804[3/] X-ray unit was used. Seeds were given a 2-minute exposure of 14 KVP at a distance of 22 inches. Following x-raying, each group of 100 seeds was placed in a pint-size plastic freezer bag along with a label to maintain identity. Each bag was filled with tap water and the seeds were allowed to soak at room temperature for 24 hours. The water was drained and the seeds were stratified

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[2/] Orthocide 75 is a product of Chevron Chemical Co., Ortho Division. Active ingredient is captan, 75% by weight.

[3/] Field Emission Corp., Melrose Ave. at Linke, McMinnville, Oregon 97128.

within the plastic bags at 5° C for 4 weeks. Following stratification, the seeds were surface dried by blotting off the excess water and air dried for 1 hour. Orthocide 75 was then applied by shaking the seeds in a jar containing the powdered fungicide until the seed coats were thoroughly covered.

The seeds were packaged and shipped to the nurseries in a styrofoam ice chest containing ice. Most shipments arrived at their destinations within 24 hours after shipment; however, the shipment to Canada was 8 days in transit. All nurseries sowed the seed in May, immediately or very shortly after receipt.

Each nursery was asked to record emergence every 7 days through 56 days after sowing. Survival counts were recorded 3, 6, 9, and 12 months after sowing. Management practices were left to the individual nurseries, although they were asked to describe nursery practices at the conclusion of the study.

Fungicide Toxicity to Seed Germination

Seeds of lot 7 were soaked, dried, stratified and dusted with the fungicides Arasan 50[4/], Orthocide 75, and Benlate[5/], according

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[4/] Arasan 50 is a product of E. I. Du Pont De Nemours & Co. (Inc.). Industrial and Biochemicals Dept., Wilmington, Del. Active ingredient is 50% thiram.

[5/] Benlate is also a product of E. I. Du Pont De Nemours & Co. (Inc.). Active ingredient is 50% benomyl.

to the seven treatments described in Table 2. Each of the seven treatments included four groups of seeds--three dusted with the three fungicides and one which received no fungicide.

The fungicides were applied to excess by shaking the seeds in a container with the fungicide. No attempt was made to determine the rate of fungicide application.

Seed soaks before stratification were conducted in tap water at room temperature. Drying was accomplished by draining the seeds and placing them on blotting paper for the specified length of time at room temperature.

Stratification was accomplished by placing the soaked seeds in plastic bags and keeping them at 5° C for 4 weeks.

Moisture contents were determined for unsoaked seeds and for seeds receiving 2-hour and 3-day drying periods after stratification. Moisture contents were determined on a wet weight basis after drying the seeds for 24 hours in a forced-air oven at 85° C.

Germination tests were conducted at 20-30° C with light, using three replicates of 50 seeds. Germination counts were conducted every 7 days and the tests were concluded after 42 days.

The germination data were analyzed by a two-factor analysis of variance. Factor 1 was fungicides, including the three fungicides and a check receiving no fungicide. Factor 2 was treatments, including treatments 1 through 7 seen in Table 2.

Table 2. Description of treatments in fungicide study.

Treatment #	Treatment
1	Not soaked, fungicides applied w/o stratification
2	Soaked, dried 2-hours, fungicides applied w/o stratification
3.	Soaked, dried 2-hours, fungicides applied pre-stratification
4	Soaked, dried 2-hours, fungicides applied post-stratification
5	Soaked, dried 3-days, fungicides applied w/o stratification
6	Soaked, dried 3-days, fungicides applied pre-stratification
7	Soaked, dried 3-days, fungicides applied post-stratification

Optimum Storage Conditions for Noble Fir Seeds

The two seed lots used in this study were provided by the Brown Seed Company, Vancouver, Washington and are listed in Table 1 as lots 7 and 8. Additional information about these lots is recorded in Table 3.

The moisture content of each lot was adjusted by placing the seeds in humidity chambers (Figure 1) over saturated salt solutions at 20° C. The salts used were LiCl, $CaCl_2 \cdot 2H_2O$, KNO_2, NaBr, $NaClO_3$ and $ZnSO_4 \cdot 7H_2O$. The relative humidities produced by the saturated salt solutions at 20° C are listed in the Handbook of Chemistry and Physics (Hodgman, Weast and Selby, 1959) and in

Table 3. Description of the noble fir seed lots used in the storage study.

Lot #	Date of collection	Total clean seed	Initial % moisture	T. Z.	Germination		Percent filled seed by x-ray	Storage at 5° C prior to testing
					Chill	No chill		
7	Sept 29 '70	11-3/4#	8.27	56	54	52	70	2 months
8	Oct 13 '70	8-1/3#	10.39	48	47	53	53	4 months

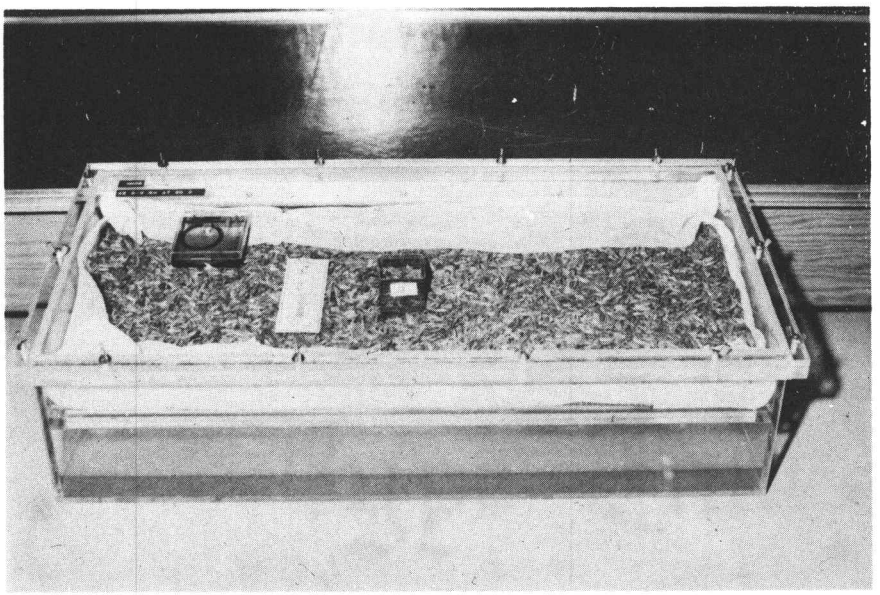

Figure 1. Humidity chamber used to adjust the seed moisture over saturated salt solutions at 20°C.

the International Critical Tables (Spencer, 1926) as 15, 30, 45, 58, 75 and 90% respectively. After 29 days the moisture content of the seeds had come to equilibrium with the relative humidity in the chambers. This method has been used to adjust the moisture of other kinds of seeds (Pixton and Warburton, 1968).

Following adjustment of moisture content, the seeds were placed in sealed quart canning jars and stored at -18, 5 and 20° C. Moisture tests were conducted at the time of storage and periodically thereafter. Seed moisture content was calculated on a wet weight basis after drying the seeds for 24 hours at 85° C.

The germination of the seeds was tested after 0, 6, 12 and 24 months storage. Four replicates of 50 seeds were chilled 4-weeks at 5° C before placing into a 20-30° C germinator with light. Another four replicates were germinated without prechilling. Germination counts were conducted every 7 days and the tests were concluded after 42 days.

A statistical analysis was conducted on the no-chill germination data using a four factor analysis of variance. Factor 1 was lots, conisting of two levels (lot 7 and 8). Factor 2 was seed moisture content, consisting of 6 levels (4, 6, 8, 9, 12 and 17%). Factor 3 was storage temperature, consisting of 3 levels (-18, 5 and 20° C). Factor 4 was length of storage, consisting of 3 levels (6, 12 and 24 months).

Statistical Analyses

Experiments subjected to statistical analysis were designed and analyzed following procedures outlined by Steel and Torrie (1960).

Experimental design was completely randomized and the data were analyzed by analysis of variance using a Control Data Corporation 3300 computer.

Duncan's new multiple range test (1% level) was used to determine differences among treatment means.

RESULTS AND DISCUSSION

Methods for Improving the Laboratory Germination of Noble Fir Seeds

Germination Temperature

Effect of Temperature on Total Germination. Maximum germination percentages achieved by each lot at each temperature are presented in Table 4.

Several temperature-germination relationships are evident from the table. Germination of Abies seeds occurred over a range of high, low, constant and alternating temperatures. Often, more than one temperature produced near maximum germination responses for a single lot. When germination means were averaged over all 12 lots, temperatures were ranked according to their ability to produce maximum germination. Highest germination occurred at 5° C. The two temperatures 20 and 5-30° C, ranked second. The two officially recommended temperatures of 20-30 and 25° C ranked third and fourth, followed by 15 and 30° C.

A significant lot X temperature interaction was found by analysis of variance (Appendix Table 1). For lots 2, 3, 10 and 12, 5° C was clearly the superior germination temperature (Table 4). Other lots, however, achieved maximum germination at another

Table 4. Effect of temperature on the germination percentages of 12 Abies seed lots.

Lot #	Germination temperature, °C						
	20-30	5-30	30	25	20	15	5
1	77	86	54	79	85	47	79
2	1	10	0	2	8	4	41
3	0	2	0	0	1	0	24
4	2	2	0	1	0	0	1
5	29	35	22	28	17	17	40
6	13	12	5	12	19	6	24
7	39	35	39	48	41	45	51
8	50	36	34	40	36	37	49
9	44	63	0	29	53	58	61
10	13	11	6	9	21	0	33
11	10	5	4	9	15	1	20
12	8	10	0	3	13	11	21
Average*	24c	25b	14f	21d	26b	19e	37a

*Those means followed by like letters are not significantly different at the 1% level of probability.

temperature. Lots 1, 4, 8 and 9 germinated highest at 5-30 and 20-30° C, but it was determined that the germination of these lots at 5° C was within tolerance of maximum germination according to Association of Official Seed Analysts tolerance tables (Association of Official Seed Analysts, 1970).

Table 5 lists those temperatures producing maximum germination and the time required for total germination to be achieved for each lot. Germination at 5° C was much slower than at the other temperatures producing maximum germination of each lot.

Germination patterns of four lots germinated at 5° C are compared in Figure 2. No germination occurred before 7 weeks, while the most rapid germination occurred between 10 and 18 weeks. This same general pattern existed for all 12 lots.

Seeds germinated at constant 30° C germinated quite rapidly but had the highest incidence of mold. Increased incidence of mold observed at high temperatures may be a reason for poorer germination at 20-30 than at 5° C. Vabre-Durrieu reported similar observations when he reported that high temperatures increased the speed of germination of Abies and increased mold growth (Edwards, 1962). The beneficial effect of 5° C germination may be due to an unfavorable environment for mold growth at that temperature.

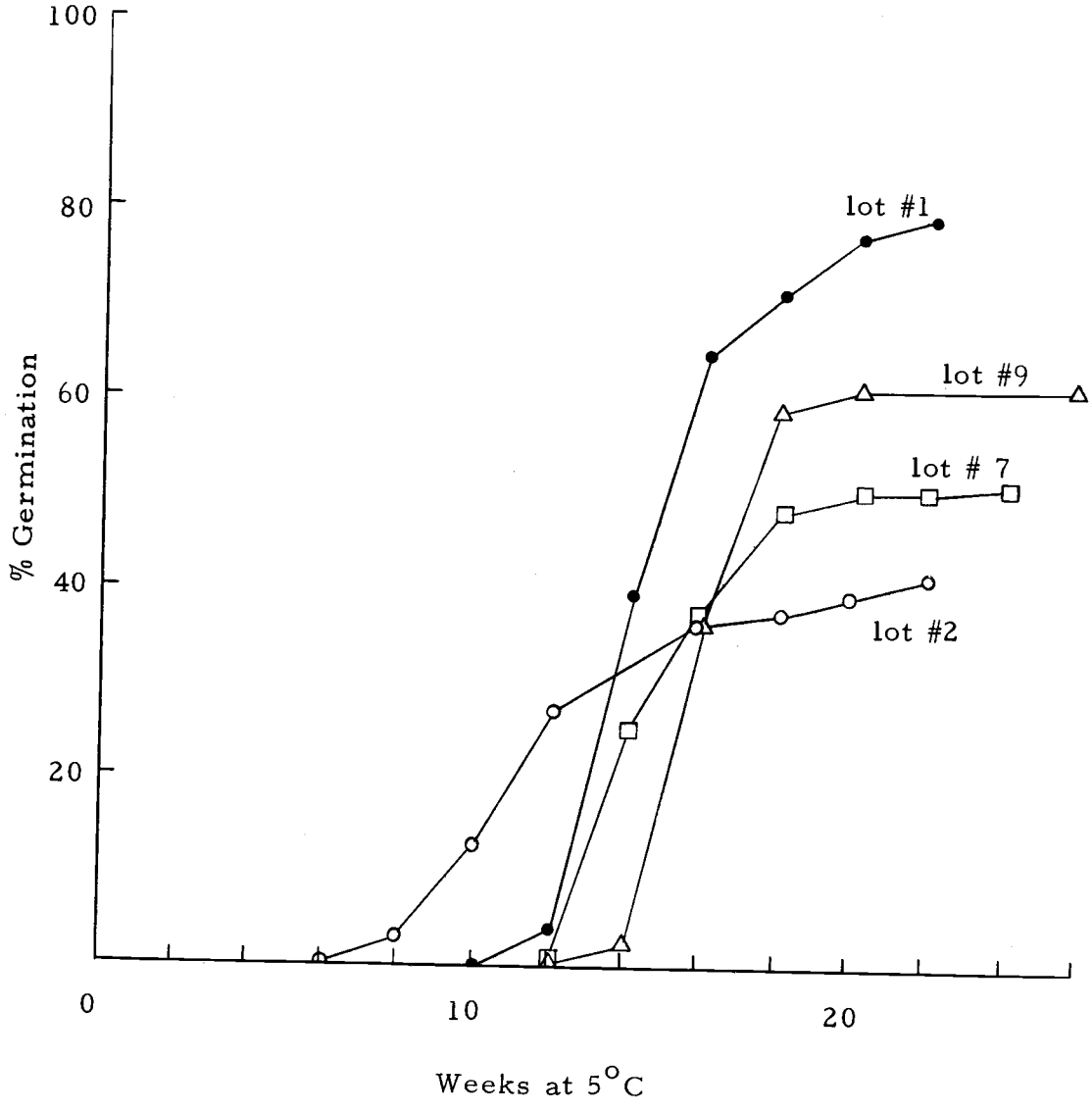

Figure 2. Seed germination patters of four selected lots of Abies germinated at 5°C.

Table 5. Temperatures producing maximum germination percentages for 12 Abies seed lots and the time required to complete the test at each temperature.

Lot #	Maximum germination %	Temperature producing maximum germination °C	Weeks to complete test
1	86	5-30	5
2	41	5	22
3	24	5	32
4	2	20-30	2
5	40	5	29
6	24	5	30
7	51	5	24
8	50	20-30	7
9	63	5-30	11
10	33	5	23
11	20	5	25
12	21	5	24

Effect of Temperature on Rate of Germination. Germination rate values for each lot at each temperature are listed in Table 6.

Alternating 20-30° C produced the highest average germination rate followed by 25, 30, 5-30, 20, 5 and 15° C in descending order. Germination rates produced at 30 and 5-30° C were not significantly different, nor were those produced at 20 and 5° C.

Germination rate is recognized as a useful tool to measure the effect of temperature (or other variable) on germination only

Table 6. Effect of temperature on the germination rate values[a/] of 12 Abies seed lots.

Lot #	Germination temperature, °C						
	20-30	5-30	30	25	20	15	5
1	32.14	22.23	24.67	29.35	6.27	3.04	5.41
2	0.00	3.33	0.00	0.75	2.57	1.13	3.50
3	0.00	0.41	0.00	0.00	0.04	0.00	1.15
4	0.75	0.40	0.00	0.50	0.00	0.00	0.06
5	12.04	7.28	10.00	10.36	3.74	2.59	2.56
6	5.88	2.51	1.67	3.83	3.33	1.23	1.92
7	13.63	8.47	15.50	13.51	5.44	2.00	3.33
8	17.89	9.06	14.75	13.55	5.66	2.43	3.29
9	9.73	10.22	0.00	5.32	4.34	2.62	3.74
10	3.24	2.37	2.83	2.77	2.06	0.00	2.03
11	2.54	0.74	1.25	2.25	1.24	0.17	1.06
12	1.95	1.99	0.00	0.66	1.54	0.67	1.07
Average*	8.36a	5.75c	5.89c	6.91b	3.02d	1.32e	2.43d

[a/] The larger values reflect faster germination.

*Those means followed by like letters are not significantly different at the 1% level of probability.

when seed of equal germination is used. The average germination rate values in Table 6 reflect germination rates of seed lots with differing maximum germination and are intended only for a general comparison. Strict use of germination rate should be limited to evaluating each lot individually.

Lot X temperature interactions were highly significant (Appendix Table 2), indicating that the highest germination rate acheived varied between lots depending on temperature. Seven of 12 lots achieved highest germination rates at 20-30° C. Other temperatures producing the highest germination rates for individual lots were 5-30, 30 and 5° C. Often the differences between the germination rate values for a single lot were small and insignificant. Generally, the warmer temperatures produced the highest germination rates. Highest germination rate was never produced by 20 and 15° C.

Table 7 lists the temperatures producing maximum germination, highest germination rate and their corresponding germination percentages. Often maximum germination was not obtained by the temperature producing the highest germination rate. For example, the maximum germination of lot 5 was 40%, occurring at 5° C. The highest germination rate for lot 5 was produced by 20-30° C, a temperature which only produced a 29% germination.

When evaluating temperatures for seed testing, maximum germination and speed of germination (germination rate) are

Table 7. Temperatures producing maximum germination, highest germination rate and the corresponding germination percentages for 12 Abies seed lots.

	Maximum germination		Highest germ. rate	
Lot #	Temp., °C	% germ.	Temp., °C	% germ.
1	5-30	86	20-30	77
2	5	41	5	41
3	5	24	5	24
4	20-30	2	20-30	2
5	5	40	20-30	29
6	5	24	20-30	13
7	5	51	30	39
8	20-30	50	20-30	50
9	5-30	63	5-30	63
10	5	33	20-30	13
11	5	20	20-30	10
12	5	21	5-30	10

important requirements. A temperature which provides rapid germination results is not acceptable if maximum germination is not achieved. The data in this study show that the temperatures which produced the highest germination rates did not usually produce maximum germination.

Length of Prechill

The effect of length of prechill on the laboratory germination of noble fir seeds is shown in Tables 8 through 13.

The speed of germination, as evidenced by the germination count at 7 days, generally increased with increasing exposure to prechill. Germination occurred during the prechill treatment, but usually not before 12 weeks. Lot 6 germinated in prechill as early as 8 weeks, but substantial germination did not occur until 12 weeks. These data support the findings reported earlier, that most germination at 5° C occurred between 10 and 18 weeks.

An analysis of variance (Appendix Table 3) showed that there were significant differences in germination percentages after different lengths of prechill. These are shown in Table 14. A 20 week prechill gave the highest overall germination for the six lots studied, followed by 18 and 16 week periods.

The significant lot X length of prechill interaction (Appendix Table 3) indicates that some lots responded differently to varying

Table 8. Germination percent of noble fir seed lot 1 following exposure to varying lengths of prechill.

Weeks prechill	0	2	4	6	8	10	12	14	16	18	20	22	24
7 day count	0	10	22	59	74	76	92	72	77	92	--	86	--
14 day count	58	82	82	88	91	85	--	73	78	--	--	87	--
21 day count	77	87	87	89	92	87	--	73	78	--	--	--	--
28 day count	80	87	87	89	92	--	--	73	78	--	--	--	--
35 day count	--	87	87	89	--	--	--	73	78	--	--	--	--
42 day count	--	--	--	89	--	--	--	73	--	--	--	--	--
Percent total germination	80	87	87	89	92	87	92	73	78	92	93	87	93
Percent firm seed	1	0	0	0	0	0	0	0	0	0	0	0	0
Percent germ. in prechill	0	0	0	0	0	0	0	10	65	88	93	86	93

Table 9. Germination percent of noble fir seed lot 2 following exposure to varying lengths of prechill.

Weeks prechill	0	2	4	6	8	10	12	14	16	18	20	22	24
7 day count	0	3	7	19	13	20	26	20	19	38	46	45	0
14 day count	0	8	7	20	13	20	26	20	19	38	46	45	0
21 day count	0	9	7	20	13	20	26	20	19	38	46	45	--
28 day count	0	9	7	20	13	20	26	20	19	38	46	45	--
35 day count	-	9	7	20	13	20	26	20	19	38	46	--	--
42 day count	-	-	--	20	13	20	26	20	19	38	46	--	--
Percent total germination	0	9	7	20	13	20	26	20	19	38	46	45	0
Percent firm seed	1	0	0	0	0	0	0	0	0	0	0	0	0
Percent germ. in prechill	0	0	0	0	0	0	26	20	19	38	46	45	0

Table 10. Germination percent of noble fir and lot 3 following exposure to varying lengths of prechill.

Weeks prechill	0	2	4	6	8	10	12	14	16	18	20	22	24
7 day count	0	0	0	2	2	1	2	2	7	10	14	13	8
14 day count	0	3	13	3	2	4	3	10	9	13	15	--	--
21 day count	1	4	15	3	2	4	3	11	9	13	15	--	--
28 day count	1	4	15	3	2	4	3	11	9	13	15	--	--
35 day count	1	4	15	3	2	4	3	11	9	13	--	--	--
42 day count	--	4	15	3	2	4	3	11	9	13	--	--	--
Percent total germination	1	4	15	3	2	4	3	11	9	13	15	13	8
Percent firm seed	0	1	2	0	0	3	3	4	0	0	0	0	0
Percent germ. in prechill	0	0	0	0	0	0	0	0	0	0	3	9	8

Table 11. Germination percent of noble fir and lot 4 following exposure to varying lengths of prechill.

Weeks prechill	0	2	4	6	8	10	12	14	16	18	20	22	24
7 day count	0	0	0	1	0	0	1	1	0	3	3	5	2
14 day count	1	1	0	1	1	0	1	1	0	3	3	5	--
21 day count	1	1	0	1	1	0	1	1	0	3	3	--	--
28 day count	1	1	0	1	1	0	1	1	0	3	3	--	--
35 day count	1	1	0	1	1	0	1	1	0	3	--	--	--
42 day count	1	1	0	1	1	0	1	1	0	3	--	--	--
Percent total germination	1	1	0	1	1	0	1	1	0	3	3	5	2
Percent firm seed	0	0	0	0	0	0	0	0	0	0	0	0	0
Percent germ. in prechill	0	0	0	0	0	0	0	1	0	3	3	5	2

Table 12. Germination percent of noble fir and lot 5 following exposure to varying lengths of prechill.

Weeks prechill	0	2	4	6	8	10	12	14	16	18	20	22	24
7 day count	0	3	2	19	19	21	18	27	35	36	45	22	31
14 day count	23	20	29	25	24	22	23	30	39	36	45	22	31
21 day count	44	27	30	26	24	22	23	30	39	37	45	22	--
28 day count	44	27	30	26	46	22	23	30	39	37	45	22	--
35 day count	45	27	30	26	46	22	23	30	39	37	45	--	--
42 day count	--	27	30	26	46	22	23	30	39	37	45	--	--
Percent total germination	45	27	30	26	46	22	23	30	39	37	45	22	31
Percent firm seed	3	0	0	0	1	0	0	2	0	0	0	0	0
Percent germ. in prechill	0	0	0	0	0	0	0	10	21	30	43	22	31

Table 13. Germination percent of noble fir and lot 6 following exposure to varying lengths of prechill.

Weeks prechill	0	2	4	6	8	10	12	14	16	18	20	22	24
7 day count	0	0	10	8	7	15	17	18	19	21	23	25	23
14 day count	7	11	14	9	7	16	17	19	19	21	--	--	--
21 day count	12	12	15	9	7	16	17	19	19	--	--	--	--
28 day count	12	12	15	9	7	16	17	19	--	--	--	--	--
35 day count	12	12	15	9	7	16	17	19	--	--	--	--	--
42 day count	12	12	15	9	7	16	17	--	--	--	--	--	--
Percent total germination	12	12	15	9	7	16	17	19	19	21	23	25	23
Percent firm seed	0	0	0	0	0	0	0	0	0	0	0	0	0
Percent germ. in prechill	0	0	0	0	3	2	9	15	19	21	23	25	23

lengths of prechill. Several lots germinated equally well at several prechill exposures. Lot 1 was relatively indifferent to length of prechill, but lot 2 benefited by prolonged prechill periods. This compliments the finds of Jensen (1964) who found that some lots of A. concolor benefited by prechilling for 4 weeks prior to germination, while the germination of others was reduced.

Table 14. Average germination percentages following exposure of six noble fir seed lots to varying lengths of prechill.

Length of prechill (weeks)	Average germination* (percent)
20	37.33a
18	33.83b
16	29.00c
12	26.83d
8	26.67d
24	26.08d, e
14	25.42d, e
4	25.42d, e
10	24.75e, f
6	24.33e, f
22	23.58f
2	23.08f
0	22.91f

*Those means followed by like letters are not significantly different at the 1% level of probability.

The overall benefit of prechilling was also noted by Roe (1948). He reported that balsam fir required a 90-day prechill period for complete germination to occur within 60 days. He also noted that the rate of germination increased with increasing prechill periods.

Low temperature germination (i. e., in prechill) reported here confirms similar reports by others (Edwards, 1969; Franklin and Kreuger, 1968; Stein, 1951).

Some lots had decayed seedlings present after prolonged prechill periods, meaning that germination had occurred in prechill and that decay had overcome the seedlings. Because of this, it would appear that noble fir should not be prechilled longer than 12 weeks prior to the germination test. This also has obvious implications in the nursery industry. MacGillivray (1955) concluded that Abies balsamea seeds could be held in stratification for 14 months, allowing a nurseryman to carry over planting stock which was not used. Such a practice looks hazardous in light of data reported here indicating that noble fir germinates at 5° C after 12 weeks.

These data point out the difficulty of recommending a single period of prechill for laboratory germination of noble fir seed. Most laboratories currently report results of germination after a 3 or 4 week prechill as well as germination results of seed receiving no prechill. Neither test results may be correct. The alternatives

would be to conduct multiple tests using different lengths of prechill or to germinate the seeds at 5° C.

Growth Regulators

Germination percentages after soaking noble fir seeds in growth regulators are shown in Table 15.

The 20-hour soak period, averaged over growth regulators and germination temperatures, produced higher germination than the 40-hour soak. However, there was a significant interaction between germination temperature and length of soak (Appendix Table 4). Seeds soaked for 40 hours produced the highest germination at the suboptimum temperature of 15° C. When seeds were germinated at 5-30° C, the 20-hour soak produced the highest germination.

Table 16 lists the germination of seeds soaked in each solution, averaged over length of soak and temperature. Soaking in GA_3, water, thiourea and KNO_3 produced the highest germination. GA_3 and water soaks both produced significantly higher germination than succinic acid or benzyladenine soaks.

Although there was not a significant solution X temperature interaction, ranking of the solutions according to their ability to produce germination responses was different depending on the germination temperature. At 15° C seeds germinated highest after soaking in GA_3, followed by KNO_3, H_2O, thiourea, succinic acid and

Table 15. Germination of noble fir seed lot 1 at two temperatures after soaking for 20 and 40 hours in growth regulators.

Length of soak (hr.)	Germination temperature													Length of soak average g/
	15° C							5-30° C						
	GA_3 a/	Thio b/	Benz c/	Succ d/	KNO_3 e/	H_2O f/	$\bar{X}$	GA_3	Thio	Benz	Succ	KNO_3	H_2O	$\bar{X}$
20	6.5	0.5	3.0	2.5	5.5	3.0	3.5	85.0	79.0	77.5	81.5	85.0	90.5	83.0	43.29a
40	15.5	7.5	2.0	4.0	10.0	6.0	7.5	74.0	77.0	59.0	64.0	63.0	69.5	67.7	37.62b
Average	11.0	4.0	2.5	3.2	7.7	4.5	5.5	79.5	78.0	68.2	72.7	74.0	80.0	75.4	

a/ Gibberellic acid

b/ Thiourea

c/ Benzyladenine

d/ Succinic acid

e/ Potassium nitrate

f/ Tap water

g/ Those means followed by like letters are not significantly different at the 1% level of probability.

benzladenine. At 5-30° C, seeds germinated highest after soaking in H_2O, followed by GA_3, thiourea, KNO_3, succinic acid and benzyladenine. Succinic acid and benzladenine maintained their low ranking regardless of the germination temperature.

The various soak treatments did not produce higher germination results than those obtained for this lot following normal laboratory testing procedures (Table 1).

Table 16. Germination of noble fir seed lot 1, averaged over germination temperatures and length of soak, after soaking in growth regulators.

Soak solution	% Germination*
GA_3 (200 ppm)	45a
H_2O	42a
Thiourea (5.63 ppm)	41a, b
KNO_3 (200 ppm)	41a, b
Succinic acid (16.8 ppm)	38b
Benzyladenine (5.63 ppm)	35b

*Those means followed by like letters are not significantly different at the 1% level of probability.

Seed Coat Chipping

Speed of germination was accelerated by removing the seed coat from the root end of noble fir seeds prior to germination

(Table 17). The overall germination percentage, however, was not affected.

Table 17. Effect of seed coat chipping on the germination of noble fir seed lot 1 at 20-30° C.

Germination counts (days)	Check		Chipped	
	No chill	4-week chill	No chill	4-week chill
7	0	23	7	86
14	59	83	87	87
21	78	87	87	87
28	80	87	87	87
35	80	87	87	87

Maximum germination was obtained within 14 days without prechilling. Edwards (1969) reported similar results. He felt that chipping might allow more rapid uptake of water and facilitate gas exchange, but his investigations proved that this was not so. He found that the seed coat did not restrict water uptake. When he chipped the seed coat at various locations other than the micropylar end, the speed of germination did not increase. Further, he found that embryo dormancy was not involved in the delayed germination of noble fir, because excised embryos from stratified and unstratified seeds grew equally well. Edwards concluded that dormancy of noble fir was due to seed coat restriction of the embryo.

There are shortcomings to this rapid method for germinating noble fir seeds. The chipping process is time consuming and extreme care must be exercised not to harm the radicle when chipping the seed coat. Improperly conducted tests might result in a lower germination because of an increased number of abnormal seedlings.

This method, however, has vast potential in seed testing because it aleviates the need for prechilling and shortens the testing period. Additional testing of a variety of samples needs to be done in order to insure that the method works for all samples.

Noble Fir Seedling Emergence and Survival

Seedling Emergence

Seedling emergence of four seed lots at five nurseries is shown in Table 18. Seedling emergence, when averaged over lots and treatments, ranged from 22 to 8% at the different nurseries. These differences were significant (Appendix Table 5).

Seedling emergence was dependent upon the conditions prevailing at the nursery where the seed lots were planted. It was not clear from the descriptions of nursery practices why emergence varied so greatly. Soil temperature and moisture during germination, planting depth and presence of soil-borne pathogens are a few

conditions which could influence seedling emergence and which may be controlled by nursery practices. The lowest seedling emergence occurred at nursery B, possibly due to seed deterioration during the delay in transit of the seeds.

Table 18. Percentage seedling emergence of four treated and untreated noble fir seed lots at five nurseries.

	Untreated lots					Treated lots*					
Nursery	1	2	3	5	$\bar{X}$	1	2	3	5	$\bar{X}$	Nursery average**
A	76	19	10	33	35	22	3	1	15	10	22a
D	58	12	8	31	27	33	5	2	13	13	20a, b
C	46	13	8	29	24	24	3	1	9	9	16b, c
E	41	11	3	17	18	25	1	0	5	8	12c, d
B	23	5	4	10	11	9	2	4	7	6	8d
Average	49	12	7	24	23	23	3	2	10	9	

* Seeds of these lots were treated with the fungicide Orthocide 75.

** Seedling emergence percentages with common letters did not differ significantly at the 1% level of probability.

No single laboratory test could have predicted the emergence at all five nurseries, because the emergence was not the same at all locations. This fact is recognized by many conifer nurseries. To solve the problem, they have each developed a factor which is applied to laboratory germination test results to help predict stand density. This factor varies between nurseries, due to the uniqueness

of cultural and environmental conditions existing between them.

The percent seedling emergence of untreated lots was higher than for treated lots. When all lots were averaged over nursery locations, seed lots treated with Orthocide 75 had an average emergence of 9% compared to 23% emergence for the untreated lots--a 57% stand reduction for Orthocide 75 treated seed. Additional findings and discussion regarding the harmful effect of Orthocide 75 are presented in the section on Fungicide Toxicity to Seed Germination.

The seedling emergence for each lot at each nursery, averaged over treated and non-treated seeds, is shown in Figure 3. It is clear that the nurseries in this study, while differing significantly in their emergence results, ranked the seed lots similarly. A minor exception was nursery B, where the relative rankings of lots 2 and 3 were reversed. This relative ranking of seed lots by nurseries is similar to the ranking of the various laboratory tests (Table 19).

When comparing the various viability determinations with the average seedling emergence at all nurseries (Table 19), it is obvious that the laboratory results were considerably higher. The germination test provided the closest approximation of field performance, but it too overestimated the seedling emergence.

The problem of lack of correlation of laboratory germination results with seedling emergence in the field may be resolved by the

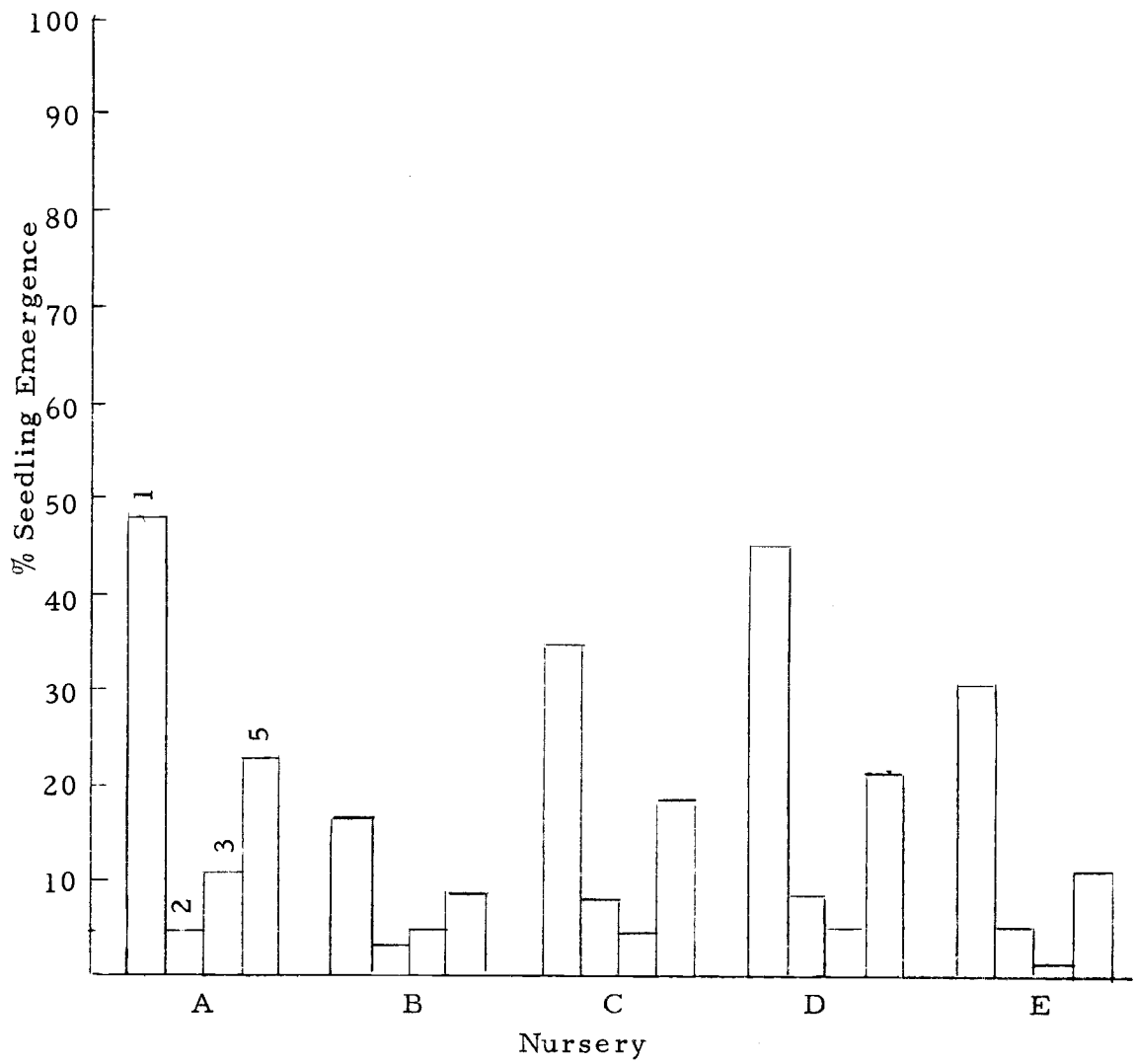

Figure 3. Seedling emergence, averaged over treated and non-treated seeds for lots 1, 2, 3 and 5 at five nursery locations.

development of a vigor test. Vigor testing measures the performance of seeds under stress conditions similar to those which they may encounter in the field. Several methods for measuring seed vigor now exist, but no test has been developed for routine laboratory vigor testing of conifer seeds.

Table 19. Comparison of laboratory test results with seedling emergence of non-treated noble fir seeds averaged over five nurseries.

	Percent viability				
Lot #	Germination*	Tetrazolium	Hydrogen peroxide	Excised embryo	Percent seedling emergence
1	84	85	93	91	49
2	4	6	21	28	12
3	8	12	21	23	7
5	38	42	57	33	24
Average	34	36	48	44	23

*Average of chill and no-chill tests.

Seedling Survival

Seedling survival data collected at 3, 6, 9 and 12 months after sowing, together with seedling emergence data collected during the first 56 days after sowing, are shown in Table 20.

The greatest seedling losses occurred within the first 3 months after sowing. Seeds treated with Orthocide 75 produced an average

Table 20. Comparison of noble fir seedling survival at 3, 6, 9 and 12 months after sowing and 56 day emergence counts.

Lot	Nursery	Treated					Non-treated				
			Percent survival					Percent survival			
		Percent emergence	3 mos.	6 mos.	9 mos.	12 mos.	Percent emergence	3 mos.	6 mos.	9 mos.	12 mos.
1	A	22	12	11	11	10	76	46	43	43	41
	B	9	3	2	1	--	23	15	13	7	--
	C	24	16	15	15	14	46	30	29	29	29
	D	33	24	24	--	19	58	48	47	--	44
	E	25	9	8	8	8	41	19	16	16	16
Average		23	13	12	9	13	49	32	30	24	33
2	A	3	2	2	2	2	19	13	13	12	12
	B	2	1	1	1	--	5	3	2	1	--
	C	3	2	2	2	2	13	8	8	8	7
	D	5	2	2	--	2	12	6	6	--	6
	E	1	0	0	0	0	11	5	4	4	4
Average		3	1	1	1	1	12	7	7	6	7
3	A	1	1	1	1	1	10	3	3	3	3
	B	4	2	1	1	--	4	2	2	1	--

Table 20. Continued.

Lot	Nursery	Treated					Non-treated				
			Percent survival					Percent survival			
		Percent emergence	3 mos.	6 mos.	9 mos.	12 mos.	Percent emergence	3 mos.	6 mos.	9 mos.	12 mos.
3	C	1	1	1	1	1	8	5	5	5	5
	D	2	1	1	--	1	8	5	5	--	4
	E	0	0	0	0	0	3	1	1	1	1
Average		2	1	1	1	1	7	3	3	3	3
	A	15	8	8	7	6	33	21	20	19	19
	B	7	5	2	1	--	10	6	5	2	--
5	C	9	6	5	5	4	29	18	18	18	17
	D	13	11	10	--	8	31	21	21	--	20
	E	5	0	0	0	0	17	5	5	5	5
Average		10	6	5	3	5	24	14	14	11	15
Grand Average		9	5	5	4	5	23	14	14	11	15

emergence of 9%. This figure declined to 5% three months after sowing, a 45% reduction. Non-treated seeds produced an average seedling emergence of 23%. This figure declined to 14% three months after sowing, a 40% reduction.

The rate of seedling decline was no faster for Orthocide 75 treated seed than for non-treeted seeds.

Seedling losses during the first three months after sowing and following emergence were found to differ between nurseries. Seedling losses during this period varied from 27 to 71% (Table 21). These kind of data further support the use, by individual nurseries, of a factor to apply to laboratory germination tests to aid in predicting stands.

Table 21. Percent noble fir seedling loss, averaged over four lots, during the first 3 months after sowing.

Nursery	Percent seedling loss	
	Treated	Non-treated
A	44	40
B	50	38
C	33	36
D	28	27
E	71	58
Average	45	40

The causes for seedling losses were not determined. Nurseries attributed losses to damping-off, hot weather during and after emergence, and insect larval feeding.

Fungicide Toxicity to Seed Germination

The germination data of noble fir seed, with and without fungicide, under varying conditions of stratification and seed moisture are shown in Table 22.

When averaged over stratification and soaking treatments, all three fungicides were found to reduce germination. Seeds receiving no fungicide germinated highest. Arasan 50 was less toxic than Orthocide 75, while Benlate produced the lowest germination.

When the germination data were averaged over fungicides, the only treatments found to be significantly lower were 1 and 2. Possible reasons for this will be discussed later.

The time of fungicide application had no effect on seed germination. The germination of seeds dusted with fungicides before or after stratification was not significantly different. Different reports have been made regarding the effect of applying fungicides pre- and post-stratification. Bloomberg and Trelawny (1970) reported delayed germination of Douglas fir seeds if treated with thiram before stratification. Cram and Vaartaja (1955) reported the toxicity of several fungicides to be greater if applied after stratification.

Table 22. Effect of fungicides, stratification and seed moisture on the germination percentage of noble fir seed lot 7.*

Fungicide	No soak	24-hour soak								Average percent germination
		2-hour dry				3-day dry				
	No-strat (Treat 1)	No-strat (Treat 2)	Pre-strat[a/] (Treat 3)	Post-strat[b/] (Treat 4)	$\bar{X}$	No-strat (Treat 5)	Pre-strat (Treat 6)	Post-strat (Treat 7)	$\bar{X}$	
Check	39	52	57	57	55	38	49	49	45	49a
Arasan 50	40	18	45	39	34	42	49	38	43	39b
Orthocide 75	17	25	24	28	26	45	44	43	44	32c
Benlate	24	12	29	32	24	39	25	37	30	28d
Treatment average	30B	27B	39A	39A	35	41A	42A	42A	42	

*Germination means followed by the same letter of the same case are not significantly different at the 1% level of probability.

[a/] Pre-stratification means that fungicides were applied to the seeds prior to stratification.

[b/] Post-stratification means that fungicides were applied to the seeds after stratification.

Seed moisture content varied depending on the length of drying. Seeds soaked 24 hours and dried for 2-hours or 3-days had moisture contents of 32 and 5% respectively. Seeds which were not soaked had a moisture content of 9%.

The length of drying (moisture content) had no effect on the germination of stratified seeds (treatments 3, 4, 6 and 7). Differences were found however, for non-stratified seeds. Seeds of treatment 2 germinated lower than seeds of treatment 5. Seeds of treatment 2 had a higher moisture content (32%) and may have been more susceptible to fungicide injury than the drier seeds of treatment 5 with a moisture content of 5%.

Soaking produced higher average germination than no-soaking, except for treatment 2. The reduced germination of seeds of treatment 1 may be due to lack of soak, rather than fungicide injury as proposed for seeds of treatment 2.

Although no significant interactions were found between fungicides and treatments (Appendix Table 6), it should be noted that there were tendencies for the fungicides to react differently to pre- and post-application of fungicides. Arasan 50 treated seeds germinated better when the fungicide was applied before stratification. Orthocide 75 treated seeds were indifferent to time of fungicide application. The germination of Benlate treated seeds was higher when the fungicide was applied after stratification.

These data support the findings of the seedling emergence study, in which emergence of Orthocide 75 treated seeds was reduced 57% over non-treated seeds.

The reasons for the toxicity of these fungicides is not clear. Some factors which should be considered in more detail are: (1) the relationship of seed moisture content and toxicity, (2) the effect of fungicide application pre- and post-stratification, and (3) relationship of rate of fungicide application and toxicity.

Optimum Storage Conditions for Noble Fir Seeds

The seed moisture contents of each lot after 29 days in the humidity chambers are shown in Figure 4. Moisture contents ranged from 4.06 to 17.08% with both seed lots achieving similar moisture levels following this equilibration period. Tests after 6 and 24 months indicated that very little change in seed moisture occurred over the 2-year storage period (Table 23).

Germination data for each of the storage conditions after 0, 6, 12 and 24 months are shown in Tables 24 and 25.

Moisture contents between 4 and 12% had no effect on the germination of noble fir seed before storage. The higher moisture level of 16 to 17%, however, reduced the germination slightly. This may have been due to rapid deterioration begun in the high humidity chambers during equilibration. Reducing the moisture content to as

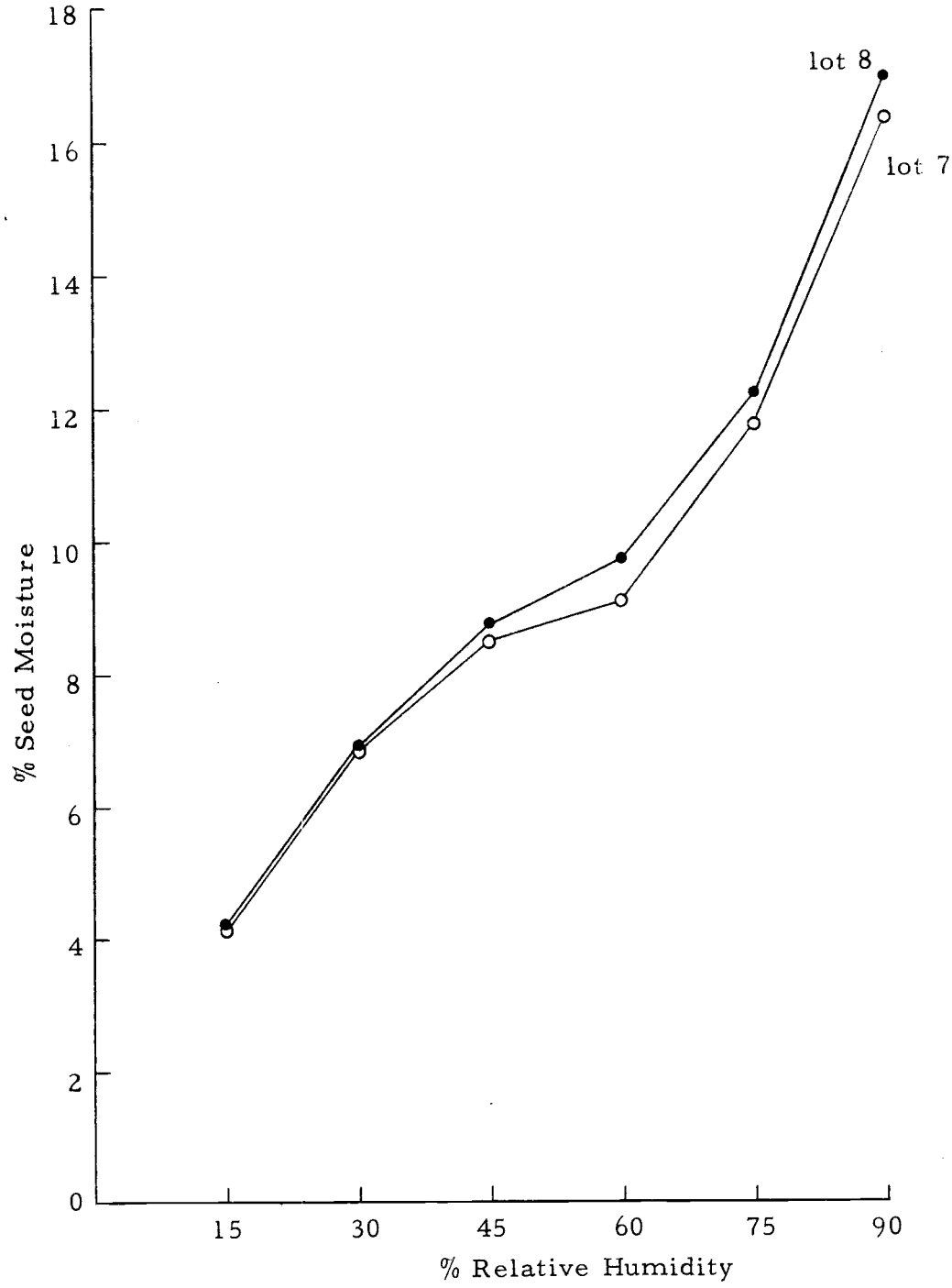

Figure 4. Seed moisture content of noble fir seed after equilibrating at different relative humidities for 29 days at 20°C.

Table 23. Seed moisture content of two noble fir seed lots after 0, 6 and 24 months storage at -18, 5, and 20° C.

Storage temperature, °C	Seed moisture content (%)					
	lot 7			lot 8		
	Original	6 mos	24 mos	Original	6 mos	24 mos
	4.06	3.96	4.20	4.19	-	3.59
	6.72	6.36	6.95	6.91	-	6.31
-18°	8.59	8.45	8.24	8.79	-	7.97
	9.02	9.43	9.39	9.84	-	9.67
	11.83	11.44	11.57	12.02	-	11.16
	16.35	15.33	14.85	17.08	-	15.09
	4.06	3.81	4.07	4.19	-	2.95
	6.72	6.97	6.19	6.91	-	6.07
5°	8.59	7.83	8.02	8.79	-	7.93
	9.02	9.47	9.47	9.84	-	8.76
	11.83	11.72	11.24	12.02	-	10.82
	16.35	15.84	16.65	17.08	-	16.23
	4.06	3.49	4.05	4.19	-	2.96
	6.72	5.83	6.72	6.91	-	6.20
20°	8.59	7.63	8.44	8.79	-	8.16
	9.02	8.74	9.45	9.84	-	9.28
	11.83	11.38	9.57	12.02	-	12.31
	16.35	15.92	16.23	17.08	-	16.21

Table 24. Effect of seed moisture content and storage temperature on the germination of noble fir lot 7 after storage for 0, 6, 12 and 24 months.

Seed moist. cont. (%)	Seed stor. temp (°C)	Percent germination							
		Before storage		6 mo. storage		12 mo. storage		24 mo. storage	
		no chill	4 wk chill	no chill	4 wk chill	no chill	4 wk chill	no chill	4 wk chill
4.06	-18			52	56	48	55	56	49
4.06	5	52	52	47	53	62	61	61	62
4.06	20			48	52	59	61	62	54
6.72	-18			53	56	51	55	55	53
6.72	5	46	55	49	52	55	51	51	51
6.72	20			46	56	48	51	50	44
8.59	-18			46	52	54	62	50	51
8.59	5	53	54	48	56	56	48	59	58
8.59	20			49	54	47	62	37	34
9.02	-18			47	54	46	53	57	50
9.02	5	58	60	49	58	56	58	50	61
9.02	20			54	59	31	22	0	0
11.83	-18			56	52	53	51	46	51
11.83	5	51	59	49	45	49	58	29	25
11.83	20			0	0	0	0	0	0
16.35	-18			54	41	33	36	16	25
16.35	5	44	45	44	50	0	0	0	0
16.35	20			0	0	0	0	0	0
Average		50.66	54.17	43.94	47.00	41.56	43.56	37.72	37.11

Table 25. Effect of seed moisture content and storage temperature on the germination of noble fir lot 8 after storage for 0, 6 and 12 months.

Seed moist. cont. (%)	Seed stor. temp (°C)	Percent germination							
		Before storage		6 mo. storage		12 mo. storage		24 mo. storage	
		no chill	4 wk chill	no chill	4 wk chill	no chill	4 wk chill	no chill	4 wk chill
4.19	-18			48	46	55	45	35	40
4.19	5	35	35	36	41	44	37	29	39
4.19	20			45	44	46	36	27	34
6.19	-18			45	47	47	45	40	37
6.19	5	41	38	35	41	40	40	35	33
6.19	20			42	38	35	34	20	30
8.79	-18			40	36	47	44	40	39
8.79	5	35	43	34	49	49	47	36	40
8.79	20			34	36	46	38	6	5
9.84	-18			39	45	48	44	40	37
9.84	5	35	37	29	35	46	42	36	31
9.84	20			32	28	0	1	1	0
12.02	-18			35	40	51	45	24	34
12.02	5	33	39	36	37	40	40	13	7
12.02	20			0	0	0	0	0	0
17.08	-18			29	24	24	18	8	14
17.08	5	27	35	12	13	0	0	0	0
17.08	20			0	0	0	0	0	0
Average		34.33	37.83	31.72	33.33	34.33	30.89	21.67	23.33

low as 4% was not harmful to the viability of the seeds, contrary to the findings of Barton (1953). She reported that both low (7 to 9%) and high (12 to 18%) seed moisture content were harmful to the germination of several Abies species.

After 6 months storage at 20° C, seeds with moisture contents of 12 to 17% lost their viability.

After 1 year storage, seeds with 16 to 17% moisture were also dead at 5° C. In addition, seeds with 9% moisture stored at 20° C were showing signs of deterioration.

After 2 years, seeds with 8 and 9% moisture stored at 20° C were declining in viability.

Germination following a 4-week prechill was higher in the early stages of storage than was germination without prechill. This is reflected in the averages shown in Tables 24 and 25. After 24 months storage the increased germination following perchill became less evident than for the fresher seeds. In this regard noble fir seems typical of many seeds which respond favorably to prechill. Older and more deteriorated seeds often germinate less, or no better than seeds receiving no prechill.

The statistical analysis conducted on the no-chill germination data is summarized in Tables 26, 27 and 28.

The effect of seed moisture on germination, when averaged over seed lots, storage periods and storage temperatures, is seen

in Table 26. These data show that the germination of seeds with moisture contents of 4, 6 and 8% was not significantly different; however, seeds with moisture contents of 12 and 17% lost viability in storage more rapidly than seeds with moisture contents of 9% and below. On the basis of these data, seed moisture contents between 4 and 8% provide the best storage for noble fir seeds.

Table 26. Effect of seed moisture content during storage on noble fir seed germination.

Seed Moisture Content (%)	Germination* (%)
4	47.58a
6	44.11a, b
8	42.97a, b
9	36.38b
12	26.52c
17	12.11d

*Those means followed by like letters are not significantly different at the 1% level of probability.

The effect of storage temperature on germination when averaged over seed lots, moisture levels and storage periods is seen in Table 27. The data indicate that -18° C and 5° C are not significantly different from one another, but that 20° C is a significantly poorer storage temperature.

The data in Tables 26 and 27 are average values which do not

show the significant interaction between storage temperature and seed moisture content (Appendix Table 7).

Table 27. Effect of three storage temperatures on noble fir seed germination when averaged over lots, moisture levels and storage periods.

Storage Temperature (° C)	Germination* (%)
-18°	43.31a
5°	37.65a
20°	23.87b

*Those means followed by like letters are not significantly different at the 1% level of probability.

Table 28 shows the effect of both temperature and seed moisture on the storage of noble fir. The germination results were obtained by averaging across lots and storage periods and clearly show the interactions between seed moisture content and storage temperature. The combination of high storage temperature and high seed moisture is not conducive to maintaining the viability of noble fir seeds. Low seed moisture content extends the temperature at which noble fir seeds can be stored. Seeds with a moisture content of 4% stored equally well at all temperatures.

The data indicate that noble fir seeds can be safely stored for 2 years at -18° C at seed moistures from 4 to 12%. At 5° C, seeds should not have a moisture content above 9%. Seeds stored at 20° C

should not have a moisture content above 4%.

Table 28. Effect of seed moisture content and storage temperature on the germination of noble fir seeds, averaged over lots and storage periods.

Seed Moisture Content (%)	Percent Germination		
	Storage Temperature (° C)		
	-18° C	5°	20°
4	48.75	46.25	47.75
6	48.41	43.91	40.00
8	46.00	46.75	36.16
9	45.75	44.08	19.33
12	43.91	35.66	0.00
17	27.08	9.25	0.00

Currently most long-term storage of noble fir seeds is at -18° C with moisture contents between 9 and 12%. The data from this study indicate safe storage under these conditions for 2 years. However, because of the added protection against high storage temperature afforded by low moisture, it seems advisable to store noble fir seeds at a moisture between 6 and 9%. The storage of seeds at lower moisture levels is probably not advisable due to the economics of drying the seeds. If it is necessary to store seeds at warmer temperatures (between 5 and 20° C), safe storage would occur with seeds at a 4% moisture level.

SUMMARY AND CONCLUSIONS

Of the methods studied to improve the germination of noble fir seeds, only seed coat chipping resulted in maximum germination within a short testing period. Removal of 1 to 2 mm of the seed coat from the radicle end of the seeds produced maximum germination within 14 days without prechill. Work on additional seed samples should be conducted because of the potential of this testing method to reduce the time required for achieving germination results.

Germination temperatures were evaluated according to their effect on total germination and rate of germination. Maximum germination occurred most often at 5° C. Germination rate increased with increasing temperatures. The highest germination rates were produced most often at 20-30° C; however, maximum germination did not usually occur at temperatures producing the maximum germination rate.

The causes for lower germination at 20-30 than at 5° C were not determined. It was thought that increased incidence of molds observed at the higher germination temperatures may be a contributing factor.

Length of prechill affected germination rate significantly. Generally, germination rate increased with increasing lengths of prechill. Lots varied in their requirements for prechill, achieving

maximum germination following different exposures to prechill. This lot X length of prechill interaction made it impossible to recommend any prechill length for obtaining maximum germination with standard seed laboratory testing procedures.

None of the growth regulator treatments increased germination over existing germination methods. Seeds soaked in growth regulators did not germinate more than seeds soaked in water. Seeds soaked for 20 hours germinated more than seeds soaked for 40 hours.

Attempts to develop a test to predict seedling emergence were negative because seedling emergence and survival varied among nursery locations. Of the viability tests studied, germination was the best indicator of field performance. All viability tests ranked the seed lots in order of seedling emergence, but not in terms of absolute percentages. It was felt that a vigor test might more closely relate to field performance.

Seedling losses occurred most noticably during the first 3 months after sowing, varying from 27 to 71% between nurseries.

Application of Orthocide 75, a common nursery practice, reduced seedling emergence of stratified seeds by 57%. Arasan 50, Orthocide 75 and Benlate all reduced laboratory germination of noble fir seeds when applied at the maximum adhering rate. Benlate was most toxic followed by Orthocide 75 and Arasan 50. Phytotoxicity was not clearly related to time of fungicide application. Arasan 50

treated seeds, however, germinated better when the fungicide was applied before stratification. Benlate treated seeds germinated higher when the fungicide was applied after stratification. The germination of Orthocide 75 treated seeds was indifferent to time of fungicide application. There was some evidence that unstratified seeds with high moisture content might be more susceptable to fungicide injury than seeds with a lower moisture content.

Storage studies indicated that noble fir seeds could be successfully stored for 2 years at -18, 5 or 20° C depending on their moisture content. The best overall storage temperature was -18° C. The viability of seeds with 12 to 17% moisture content was maintained longer at -18° C than at the other temperatures. Seeds with 4 to 9% moisture stored equally well at -18 or 5° C. Seeds with 4% moisture content stored equally well at all temperatures. These data indicate that the recommended storage moisture of 9 to 12% for noble fir could be lowered to the recommended moisture for other conifer species of the Pacific Northwest (6 to 9%) without adversely affecting the viability of these seeds in storage.

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APPENDIX

Appendix Table 1. Analysis of variance of the germination of Abies seeds as affected by temperature.

Source of variation	Degrees of freedom	Sum of squares	Mean squares	F
Lots	11	7.26057	6.60052	323.1968**
Temp (germ. temp.)	6	7.45772	1.24295	60.8617**
Lots x Temp	66	9.77684	1.48134	7.2534**
Error	84	1.71550	2.04226	
Total	167	9.15558		

**Significant difference at 1% level of probability.

Appendix Table 2. Analysis of variance of germination rate of Abies seeds as affected by germination temperature.

Source of variation	Degrees of freedom	Sum of squares	Mean squares	F
Lots	11	4.11762	3.74329	335.4420**
Temp (germ. temp.)	6	9.61980	1.60330	143.6741**
Lots x Temp	66	2.09196	3.16964	28.4036**
Error	84	9.37379	1.11592	
Total	167	7.26530		

**Significant difference at 1% level of probability.

Appendix Table 3. Analysis of variance of the germination of noble fir seeds as affected by varying lengths of prechill.

Source of variation	Degrees of freedom	Sum of squares	Mean squares	F
Lots	5	1.25622	2.51244	1210.0674**
Time (length of chill)	12	2.62908	2.19090	10.5521**
Lots x Time	60	7.86152	1.31025	6.3106**
Error	78	1.61950	2.07628	
Total	155	1.37732		

**Significant difference at 1% level of probability.

Appendix Table 4. Analysis of variance of the germination of noble fir seed lot 1 as affected by soaking in growth regulators for 20 and 40 hours.

Source of variation	Degrees of freedom	Sum of squares	Mean squares	F
Soln (growth regulator)	5	468.1667	93.6333	5.027**
Time (soak time)	1	385.3333	385.3333	20.689**
Temp (germ. temp)	1	58660.0833	58660.0833	3149.535**
Soln x Time	5	262.4167	52.4833	2.818
Soln x Temp	5	161.6667	32.3333	1.736
Time x Temp	1	1121.3333	1121.3333	60.206**
Soln x Time x Temp	5	91.9167	18.3833	0.987
Error	24	447.0000	18.6250	
Total	47	61597.9166		

**Significant difference at 1% level of probability.

Appendix Table 5. Analysis of variance of field emergence of noble fir seedlings as affected by nursery locations and fungicide treatment.

Source of variation	Degrees of freedom	Sum of squares	Mean squares	F
Fung	1	7357.6563	7357.6563	112.099**
Lots	3	23575.0688	7858.3563	119.727**
Nurs	4	4168.9625	1042.2406	15.879**
Fung x Lots	3	2418.8687	806.2896	12.284**
Fung x Nurs	4	1672.1875	418.0469	6.369**
Lots x Nurs	12	2870.0875	239.1740	3.644**
Fung x Lots x Nurs	12	1319.1625	109.9302	1.675
Error	120	7876.2500	65.6354	
Total	159	51258.2437		

**Significant difference at 1% level of probability.

Table 6. Analysis of variance of the germination of noble fir seed lot 7 as affected by fungicides applied under different conditions of seed moisture and stratification.

Source of variation	Degrees of freedom	Sum of squares	Mean squares	F
Fung	3	1.25403	4.18011	15.8095**
Treat	6	6.76142	1.12690	4.2620**
Fung x Treat	18	1.00404	5.57804	2.1097
Error	56	1.48066	2.64404	
Total	83	4.41489		

**Significant difference at 1% level of probability.

Table 7. Analysis of variance of the germination of noble fir seeds as affected by storage for varying lengths of time under different conditions of seed moisture and storage temperature.

Source of variation	Degrees of freedom	Sum of squares	Mean squares	F
Lots	1	7526.0416	7526.0416	298.992**
MC (Moisture content)	5	32490.0787	6498.0157	258.151**
Temp (storage temp.)	2	14400.5926	7200.2963	286.051**
Time (length of storage)	2	3190.7037	1595.3518	63.379**
Lots x MC	5	273.5972	54.7194	2.173
Lots x Temp	2	217.3333	108.6666	4.317**
Lots x Time	2	706.3333	353.1666	14.030**
MC x Temp	10	9776.6851	977.6685	38.840**
MC x Time	10	2388.0740	238.8074	9.487**
Temp x Time	4	342.3518	85.5879	3.400**
Lots x MC x Temp	10	1176.9444	117.6944	4.675**
Lots x MC x Time	10	1656.1111	165.6111	6.559**
Lots x Temp x Time	4	304.6666	76.1666	3.025**
MC x Temp x Time	20	6349.7037	317.4851	12.612**
Lots x MC x Temp x Time	20	1374.7222	68.7361	2.730**
Error	108	2718.5000	25.1713	
Total	215	84892.4398		

**Significant difference at 1% level of probability.